# Proceedings of the Eighth Annual Tropical and Subtropical Fisheries Conference of the Americas

TAMU-SG-83-112
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Texas A&M University

Sea Grant College Program

College Station, TX 77843

# PROCEEDINGS OF THE EIGHTH ANNUAL TROPICAL AND SUBTROPICAL FISHERIES CONFERENCE OF THE AMERICAS

Compiled by

Ranzell Nickelson II



The Tropical and Subtropical Fisheries Technological Society of the Americas is a professional and educational association of fishery technologists interested in the application of science to the unique problems of production, processing, packaging, distribution and utilization of tropical and subtropical fishery species.

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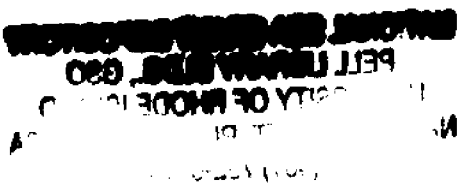

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# ACTIVITIES OF THE GULF OF MEXICO FISHERY MANAGEMENT COUNCIL

Terrance R. Leary
Gulf of Mexico Fishery Management Council
5401 West Kennedy Boulevard
Tampa, Florida 33609

When the Congress of the United States passed the Magnuson Fishery Conservation and Management Act in 1976, it extended its fishery jurisdiction to 200 miles and provided for management within this fishery conservation zone. Eight Councils were authorized to develop plans for managing the fisheries within their geographical area of authority.

Because the principal function of Council is management, we rely upon published material, and for new data needs we are dependent upon the National Marine Fisheries Service (NMFS). This presentation will outline our past activities, some of our research needs, and our utilization of the data generated by NMFS, the State conservation agencies, and other research institutions.

In 1977 the Gulf of Mexico Fishery Management Council began identifying those fisheries in need to management and developing management plans for them. For the purpose of this management program a "fishery" is defined as one or more stocks of fish which can be treated as a unit for purposes of conservation and management, and which are identified on the basis of geographical, scientific, technical, recreational, and economic characteristics.

Plans have been prepared for the following fisheries:

## STONE CRAB (Menippe mercenaria)

This is a small fishery located almost entirely in south Florida. The annual production is about two million pounds of claws from 250,000 to 300,000 traps. The plan was initiated at the request of fishermen because of conflict over fishing area between crab trap fishermen and shrimp trawlers.

Monitoring the fishery and assessing the stock is done by providing fishermen with logbooks in which they record catch and effort. NMFS maintains the catch statistics and re-evaluates the maximum sustainable yield. The Council is also sponsoring a study by a university graduate student to determine by electrophoretic analysis whether there may be substocks in the western Gulf.

The management regulations which became effective in 1979 provide for a seasonal closure to shrimping (February 15th to April 15th) in an area of high stone crab productivity; establish a closed season for taking stone crabs (May 15th to October 15th) during the spawning season; prohibit harvest of egg bearing females; allow harvest of claws seven centimeters in length, require return of declawed crabs to the water; and require shading of the crabs in the live box until they are returned to the water.

The Council is presently drafting an amendment to address a similar gear conflict problem occurring off Crystal River. Once again a seasonal zoning would separate the two groups of fishermen.

## SHRIMP (Penaeus Sp.)

The shrimp plan was implemented in May of 1981, the principal management technique used is to defer harvest of shrimp until they reach a large size in order to increase the yield per recruit. One of the key data points needed was the determination of instantaneous natural mortality of the different species of shrimp at various ages and in different locations - such as in the bays as opposed to the open Gulf.

In the case of the brown shrimp, P. aztecus, in the western Gulf, the harvest is directed at shrimp 112 mm in total length and larger. Because of the seasonal spawning and migration of this species, a closure of Gulf waters from June 1st through July 15th off the Texas coast is used to protect juvenile shrimp until they attain this size. When the season opens, all shrimp regardless of size may be retained.

State researchers monitor the size, and movement of the shrimp within the estuaries so that seasonal closure may be adjusted to conform to variable environmental conditions.

NMFS, analyzing the results of the first year's seasonal closure estimated the benefits of this measure to be the landing of an additional four million pounds of shrimp valued at $9.4 million.

The results of the second seasonal closure showed marginal benefits because most shrimp remained in the State's territorial sea and thus were not directly benefited by the FCZ closure. There is, I believe, a synergistic effect on the joint closure of adjoining state and federal waters; however, this was not quantified. Only the impact of the federal closure was measured.

The management strategy for pink shrimp, P. duorarum, in the eastern Gulf is to defer harvest until they reach a size of about 103 mm. Because of the longer spawning period of this species, the protection of the juveniles is achieved by the establishment of a permanent sanctuary in the Gulf adjacent to bay nursery areas. Shrimp grow rapidly as they move through the sanctuary and have for most part reached harvest size when they enter the area open to fishing. The population structure is not uniform, however; and NMFS researchers are finding some large shrimp in

the sanctuary and small shrimp outside. Generally, however, the sanctuary does protect the majority of the undersized shrimp in that area. We are presently modifying the plan to allow seasonable opening and closing of portions of the sanctuary based on sampling.

## SPINY LOBSTER (Panulirus argus)

The management program for the spiny lobster was implemented in April of 1982. This fishery is conducted off the southern coast of Florida, and because it occurs on the Atlantic as well as the Gulf side, the plan was developed jointly with the South Atlantic Council. The optimum yield of the fishery is about ten million pounds per year. Principal management measures include a closed season April 1st through July 25th during the primary spawning season; a minimum harvestable size limit of three-inch carapace length; return of egg bearing females, and a degradable panel in the trap to allow escapement of lobsters from lost traps.

Because of the prolonged larval stage of this species, we are not sure of the source (the locations of the principal spawners). This is an important research need. The Councils have therefore been conservative by making the management assumption that the Florida fishery is dependent on Florida spawners.

## COASTAL MIGRATORY PELAGICS

Included in this management unit are the king, Spanish, and cero mackerels, little tunny, bluefish, cobia, and the dorado. This, a joint plan with the South Atlantic Council, should be implemented at any time. It provides for minimum size limits of 12 inches for Spanish mackerel and 33 inches for cobia, both intended to increase the yield per recruit.

Another measure would allow restriction of gear to separate gear-type users (net and handline fishermen) in the event a gear conflict occurs in the king mackerel fishery. The plan also provides for an allowable catch of king mackerel not to exceed 37 million pounds with 28 million for the recreational fishery, 3.9 million for the commercial hook and line fishery, and 5.1 million for the commercial net fishery. Provision is made to close the fishery for the remainder of the year to any user group exceeding its limit.

Researchers believe that we have two stocks of king mackerel in the U.S. fishery. One stock is restricted to the Atlantic coast; the other is believed to migrate from the Atlantic coast of Florida into the Gulf of Mexico – ranging at least to Texas. We need to know more about migrations in the western and southern Gulf and determine the extent to which we share stocks of these migratory species with Mexico.

Recently, a winter fishery for king mackerel has developed off Louisiana, and production may reach 500,000 pounds this season. The range of this group of fish is unknown, but NMFS is presently tagging in the area to answer this question.

## REEF FISH

Principal fishes in this group are the snappers and groupers. The plan has been submitted and should be approved at any time. Management measures identify stressed areas where the reef fish populations are heavily exploited. In these stressed areas roller trawls, fish traps, and power heads for taking reef fish are to be prohibited.

NMFS has provided yield per recruit analysis for some species which allows us to increase the potential yield by placing a size limit on a species such as the minimum size of 12 inches which is proposed for the red snapper. We must also have data on the survival of caught and released fishes in order to determine the impact of size selection. The NMFS is also conducting studies to provide this information.

## CORAL

Also scheduled for implementation soon is the plan for coral; another joint plan with the South Atlantic Council. Stony corals and two species of sea fans would be protected from harvest. Additionally, several unique coral areas are to be designated for special protection by prohibiting within them the use of certain bottom fishing gear that could damage coral.

Other fisheries for which plans are being developed are swordfish, the billfishes including the marlins and sailfish, and the calico scallop. All are joint plans being developed by the South Atlantic Council. In the case of the scallop, the Councils decided to proceed with management on learning that numerous small scallops were being caught, landed, and discarded in order to land a few large individuals. We estimated that during December of 1981 35 million small scallops were landed and discarded each day. In 1981 scallop production was about 20 million pounds of meats, with most of the production coming from the Atlantic coast rather than the Gulf of Mexico.

Tentative plans have been prepared for several other fisheries but have not been submitted for implementation for various reasons. There is an insufficient data base to manage sharks. The groundfishes, composed mostly of croakers, cannot presently be managed without adversely affecting the shrimp fishery. Coastal herrings (sardines, anchovies, scads, bumpers, and others) are underutilized species and are not presently in need of management regulations.

The Council is presently reviewing the condition of the red drum stock following reports of large catches being made as a bycatch in the purse seine fishery for blue runners.

The Council has also been active in reviewing and commenting on various projects which could have detrimental impacts on the habitat of those species managed under the FMPs. We became actively involved in projects to restore Colorado River flow to Matagorda Bay, Texas; to stop the dumping of dredged material from Tampa Bay on coral reefs and hard

bottoms offshore; and to utilize onshore sites for spoil placement from the Corpus Christi Harbor. In the latter case, the Council held a public hearing in Corpus Christi to seek public input on alternatives to the filling of some 1,200 acres of shrimp nursery habitat in Nueces Bay.

The U.S. Army Corps of Engineers has agreed to incorporate the water diversion plan in the Colorado River project and has approved a plan for onshore spoil disposal at Corpus Christi. In Florida, a law suit brought by Manatee County, stopped the dumping on the offshore hard bottoms.

In the next three years we anticipate implementation of the remaining active plans. We will continue to watch closely the shrimp stocks to observe any parent-recruit relationship. Our Council has also identified some specific study areas. Included are, a continuous monitoring of the impact of the seasonal shrimping closure off Texas; monitoring shrimp size distribution in the Tortugas Shrimp Sanctuary; a tagging study on migration patterns of king mackerel; a size distribution study by area of the king mackerel catch; and a survey of the recreational catch of all species of fish. Monitoring of all plans and regulations will continue so that management objectives can be adjusted where needed, and regulations may be better tuned to plan objectives.

# WORLD SHRIMP PRODUCTION TRENDS AND THE U.S. IMPORT MARKET

Fred J. Prochaska, Mauro Suazo and Walter R. Keithly
Department of Food and Resource Economics
University of Florida

## INTRODUCTION

U.S. shrimp consumption increased 40 percent from approximately 226,000 metric tons (MT) in 1965 to a high of 347,000 MT in 1977 (Figure 1). Shrimp imports exceeded U.S. domestic production in twelve of the sixteen years between 1965 and 1980. Between 1965 and 1970, domestic production increased more rapidly than did imports (52 percent compared to 33 percent). During this period, exports increased nearly 150 percent. After 1970, exports leveled off. With the exception of record production in 1977, imports have increased at a greater annual rate than domestic landings. Since 1970, however, both have tended to level off in recent years.

The level of U.S. consumption is dependent on shrimp imports. Furthermore, a comparison of annual changes shows changes in imports were in the same direction of consumption changes in ten of the twelve years up to 1976. Since 1975, consumption variation has been closely associated with changes in domestic production while imports have leveled off. Future trends in the U.S. shrimp market are highly dependent on shrimp imports which, in turn, are dependent on world production and consumption levels.

The purpose of this paper is to (1) review world shrimp production trends in total and by major producers, (2) investigate U.S. shrimp supply sources and trends, (3) analyze the U.S. market for imported shrimp, and (4) draw implications with respect to future conditions in the U.S. shrimp market.

## WORLD SHRIMP PRODUCTION

World production of shrimp[1] increased from 376,000 MT in 1950 to a high of 1,699,000 MT in 1978 (Table 1). With the exception of 1975 and 1979, world shrimp production increased each year since 1958. The average rate of increase between 1950 and 1978 was approximately 47,250 MT per year. Since 1977, total production appears to have leveled out at an average of 1,650,000 MT annually.

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[1] FAO statistics contain some undetermined amount of Prawn Production.

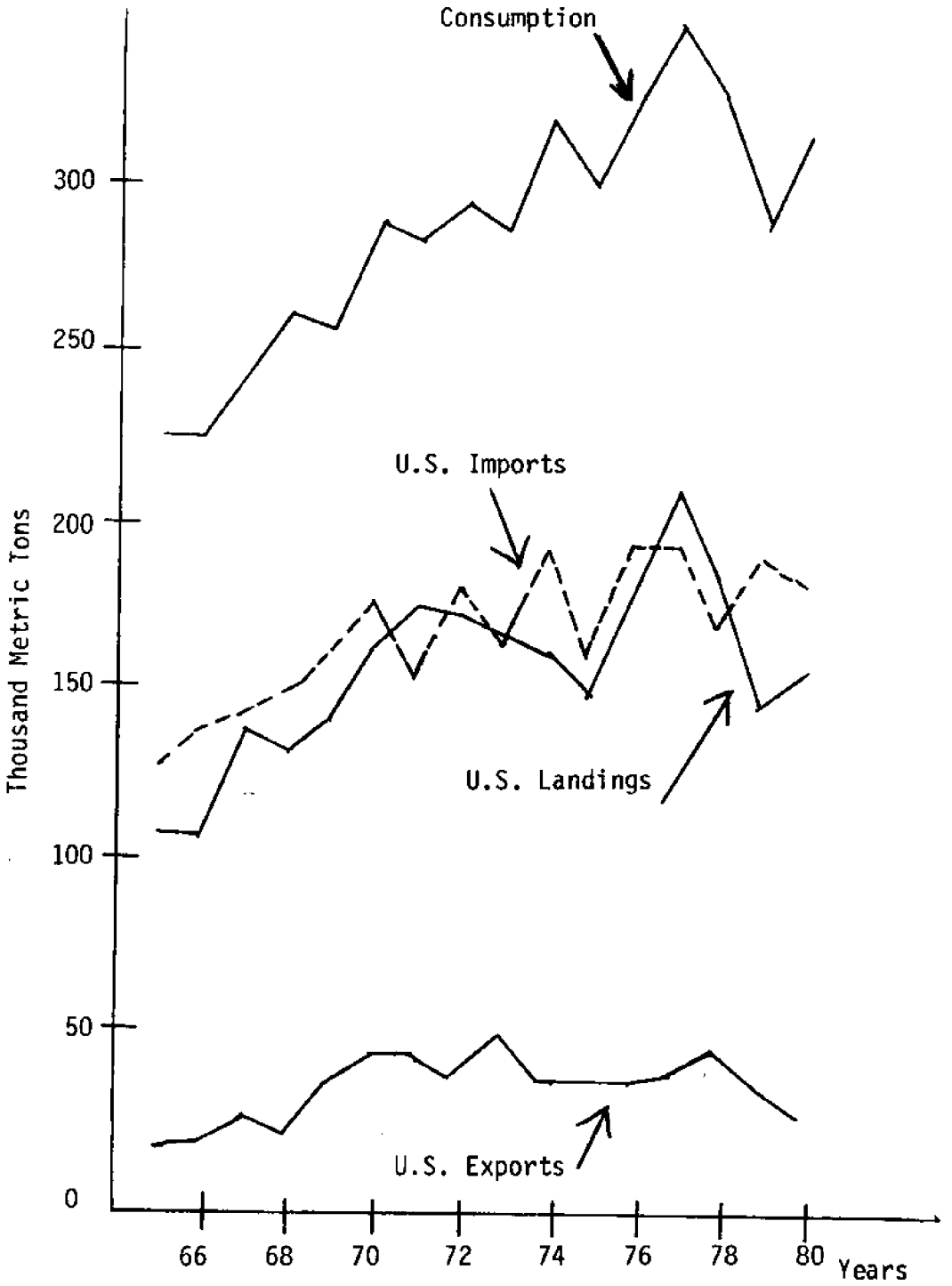


Figure 1. U.S. Shrimp production, consumption, imports and exports, 1965-80. (Derived from 5).

FAO (4) 1970 estimates of total MSY for world production is 1,492,600 MT with a range on the estimate from 1,343,340 MT to a high of 1,641,860 MT. Since the 1970 estimates were made, a small increase in MSY was estimated for some countries. Current world production appears to be within the range of estimated MSY.

World production of shrimp has been, and continues to be, highly concentrated. Comparing relative production for each fifth year for the years 1965, 1970, 1975 and 1980 shows between 48 to 51 percent of total world production is produced by the five leading producers and between 69 and 70 percent is produced by the top ten producers (Table 2). Production was somewhat more concentrated in 1960 when 67 percent was produced by the top five and 83 percent was produced by the top ten producers.

The U.S. was the leading shrimp producer in 1960, accounting for 22.5 percent of total world production (Table 2). By 1980, the world share accounted for by the U.S. declined to 9.4 percent and the U.S. dropped to third in world production. The U.S. share of world production peaked in 1953 when she accounted for 26.4 percent.

Table 1. World Shrimp and Prawn Landings (1,000 metric tons), 1950-80 (3).

| Year | MT | Year | MT | Year | MT |
|---|---|---|---|---|---|
| 1950 | 376 | 1960 | 501 | 1970 | 966 |
| 1951 | 395 | 1961 | 509 | 1971 | 1,031 |
| 1952 | 395 | 1962 | 556 | 1972 | 1,113 |
| 1953 | 447 | 1963 | 614 | 1973 | 1,260 |
| 1954 | 511 | 1964 | 690 | 1974 | 1,349 |
| 1955 | 432 | 1965 | 700 | 1975 | 1,331 |
| 1956 | 533 | 1966 | 730 | 1976 | 1,450 |
| 1957 | 518 | 1967 | 800 | 1977 | 1,672 |
| 1958 | 466 | 1968 | 830 | 1978 | 1,699 |
| 1959 | 474 | 1969 | 860 | 1979 | 1,560 |
| | | | | 1980 | 1,681 |

The 9.4 percent share in 1980 is the lowest for the 31-year period between 1950 and 1980.

Since 1973, India has been the leading world shrimp producer. India's world share was at a high in 1954 with 30.1 percent of world production. Shrimp production in India reached a maximum at 159,600 MT in 1956 and then declined until the early 1960s (3). Production then trended upward through 1974 when a high of 246,000 MT was reached. Production then became highly variable, ranging from a low of 183,100 MT in 1979 to a high of 244,400 MT in 1980. With the exception of a few years, India has accounted for between 11 percent and 14 percent of world production since 1960. Overall, India's production has kept pace with world production trends.

Table 2. Major Shrimp-Producing Countries and Shrimp Production (1,000 metric tons), 1960-80 (derived from 3).

| 1960 | | | 1965 | | | 1970 | | | 1975 | | | 1980 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Country | Vol. | % | Country | Vol. | % | Country | Vol. | % | Country | Vol. | % | Country | Vol. | % |
| U.S.A. | 112.9 | 22.5 | U.S.A. | 110.5 | 15.7 | U.S.A. | 166.7 | 17.2 | India | 237.8 | 17.9 | India | 244.5 | 14.5 |
| India | 70.6 | 14.1 | India | 77.3 | 11.0 | India | 115.2 | 12.0 | U.S.A. | 153.0 | 11.5 | Indonesia | 175.3 | 10.4 |
| Mexico | 66.8 | 13.3 | Japan | 66.8 | 9.5 | Thailand | 85.9 | 8.9 | Thailand | 110.5 | 8.3 | U.S.A. | 158.2 | 9.4 |
| Japan | 60.0 | 11.9 | Mexico | 59.6 | 8.5 | Mexico | 67.2 | 7.0 | Indonesia | 72.5 | 5.4 | China | 143.6 | 8.5 |
| Brazil | 23.4 | 4.7 | Brazil | 39.4 | 5.6 | Japan | 54.5 | 5.6 | Mexico | 70.0 | 5.2 | Thailand | 107.8 | 6.4 |
| S. Korea | 19.4 | 3.9 | Thailand | 39.2 | 5.5 | Malaysia | 47.0 | 4.9 | Japan | 69.1 | 5.1 | Malaysia | 84.0 | 5.0 |
| Pakistan | 18.0 | 3.6 | Pakistan | 32.0 | 4.6 | Brazil | 40.8 | 4.2 | Viet Nam | 62.0 | 4.6 | Brazil | 83.0 | 4.9 |
| Spain | 17.1 | 3.4 | Chile | 21.3 | 3.0 | Pakistan | 34.3 | 3.5 | Phillip. | 52.5 | 3.9 | Mexico | 77.5 | 4.6 |
| Thailand | 14.5 | 2.9 | Malaysia | 20.8 | 2.9 | Viet Nam | 33.3 | 3.4 | Brazil | 50.2 | 3.9 | Viet Nam | 62.0 | 3.7 |
| Netherl. | 12.7 | 2.5 | Viet Nam | 20.0 | 2.8 | Taiwan | 30.7 | 3.2 | Malaysia | 47.6 | 3.0 | Japan | 51.0 | 3.0 |
| TOTAL | 415.4 | 82.8 | | 486.9 | 69.1 | | 675.6 | 69.9 | | 925.2 | 68.7 | | 1,186.9 | 70.4 |

India's production is not expected to increase substantially unless fishing effort is increased from present levels, which could be an expensive task due to high fuel cost. However, the Indian government is at the present considering some sort of relief from high oil prices and is also trying to build up her fleet to 350 vessels (1).

Data have only been available for China since 1975, and Indonesia since 1971. Indonesia accounted for 10.4 percent of world production in 1980 and was second in world production behind India. China was fourth, with an 8.5 percent share of world production in 1980.

Thailand has consistently been in the top ten producing nations (Table 2). Production in Thailand generally increased throughout the 1950-80 time period with a low of 9,200 MT in 1959 and a high of 146,100 MT in 1978 (3). In relative terms, Thailand's best year was 1968 when she accounted for 9.5 percent of world production. Overexploitation of the resource due to overcapacity of the fleet is a problem that Thailand must look at and try to solve if it is to preserve the fishery (1).

Mexico and Brazil are the only two Latin American countries in the top ten in terms of world shrimp production (Table 2). Both countries have generally declined in relative importance in world production even though production within each country has increased.

Japan's shrimp production increased throughout the early 1960s to a high of 86,800 MT in 1963 but since then has declined to 51,000 MT in 1980. Japan's share of world production has declined from 14.5 percent in 1961 to 3.0 percent in 1980.

## U.S. SHRIMP IMPORTS

U.S. total shrimp imports increased consistently until the 1970s and then leveled off between 200 and 228 million pounds annually (Figure 1). Once the volume of imports began to stabilize, the price and total value of imports began to increase rapidly. Total value of shrimp imports increased from $196 million in 1971 to $724 million in 1981, an increase of 270 percent (Figure 2). This increase was due mainly to price increases.

Shell-on shrimp are the major product form imported (Figure 2). Shell-on shrimp have ranged from 51 to 73 percent of total U.S. shrimp imports. There has been no trend in volume of shell-on imports. Total value of shell-on imports increased in a parallel pattern to total value of shrimp imports increasing from $135 million in 1971 to $520 million in 1981.

North American countries are the main source of U.S. shrimp imports (Figure 3). Imports from these countries increased from around 90 million pounds in the mid 1960s to 115 million in 1979 and 1980 and have averaged over 50 percent of total U.S. imports

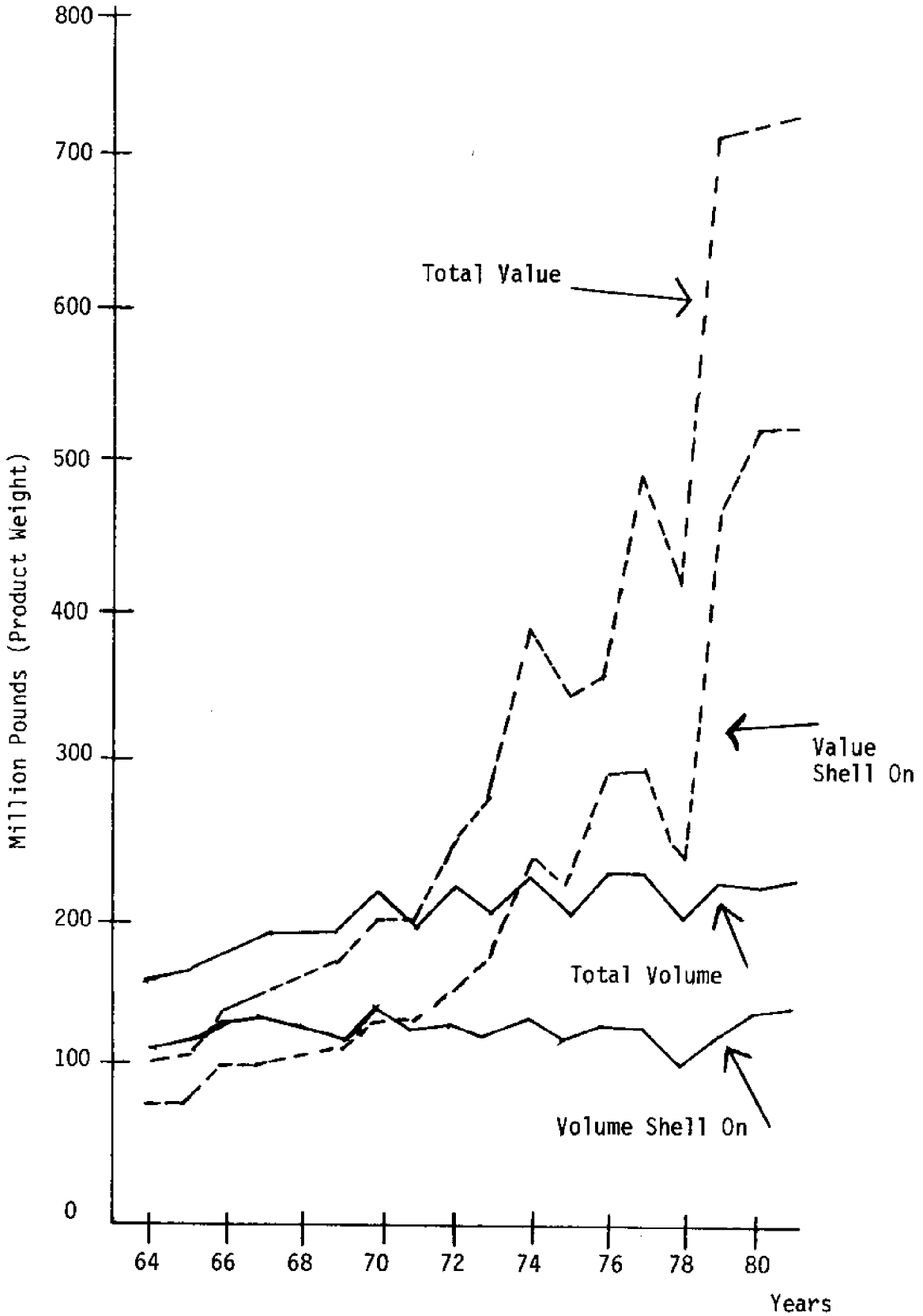


Figure 2. Value and volume of U.S. shrimp imports, 1964-81. (Derived from 6).

between 1964 and 1981. Mexico and Panama are the two principal North American exporters to the U.S. However, Central American countries have lately been securing a larger share of the U.S. market.

Imports to the U.S. from Asian countries increased dramatically between 1964 and 1978 (Figure 3). Between 1964 and 1981, Asian imports ranged from a low of 27 million pounds in 1964 to a high of 75 million pounds in 1976. Imports have been, however, highly unstable from Asian sources with variations as much as 30 million pounds (50 percent) from year to year. India, Thailand and China are the principal Asian exporters to the U.S.

Shrimp imports to the U.S. from South American countries increased from approximately 26 million pounds in 1964 to 44 million pounds in 1970 and 1972 but then declined to 28 million pounds by 1978 (Figure 3). Imports then increased to record levels of approximately 49 million pounds in 1980 and 1981. North and South America, together, generally account for between 70 to 80 percent of U.S. shrimp imports with the exception of the high Asian imports between 1974 and 1979, when their share dropped to between 64 and 70 percent. Ecuador, Brazil and Colombia are presently the principal South American exporters to the U.S.

Europe, Africa and Australia account for the remaining U.S. shrimp imports. Jointly, European imports are rather insignificant, ranging from 1.5 to 8.5 million pounds between 1964 and 1981. Imports from each of these areas are highly unstable on an annual basis.

Mexico has been, and continues to be, the leading shrimp export country to the U.S. (Table 3). Comparing five-year periods between 1965 and 1980 shows Mexico's share of U.S. imports has declined from 64.8 percent in 1960 to 34.6 percent in 1980. However, since 1970, Mexican imports have been a stable source of U.S. total shrimp imports, generally ranging from 72 to 80 million pounds annually and accounting for between 32 to 38 percent of total U.S. shrimp imports since 1965.

India was the seventh leading shrimp exporter to the U.S. in 1960 but increased to second in 1965, 1970 and 1975 with her share ranging from 7.4 to 15.3 percent of total U.S. imports in these years. India dropped to fourth in 1980 with 5.9 percent of U.S. imports. The drop in India's share of the U.S. market was due to the FDA's actions taken against Indian imports because of the presence of salmonella bacteria in some shipments. The Indian government is now trying to remove its companies from the FDA's blacklist (1). From one half to 89 percent of Indian imports have been peeled shrimp, compared to mostly shell-on shrimp from other sources.

Ecuador became the second leading exporter to the U.S.in 1980 with exports of slightly over 20 million pounds and 9.2 percent of total U.S. shrimp imports. Cultured shrimp have been one source of

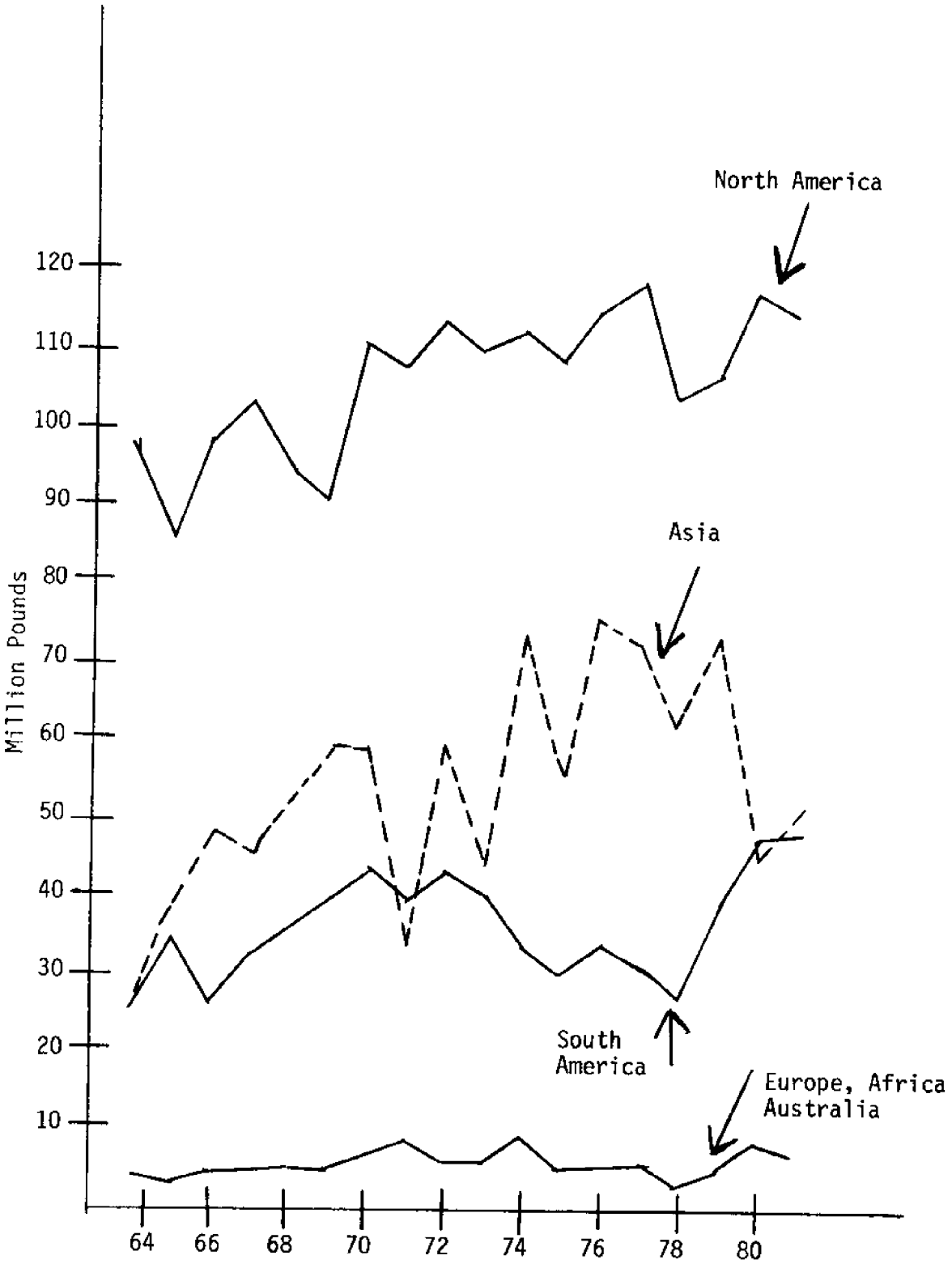


Figure 3. U.S. Shrimp imports by major production areas, 1964-81. (Product Weight). (Derived from 7).

Table 3. Principal Shrimp Supplying Countries to the United States and Shrimp Supplies (thousand pounds product weight), 1960-80 (Derived from 7).

| 1960 | | | 1965 | | | 1970 | | | 1975 | | | 1980 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Country | Vol. | %[a] | Country | Vol. | % | Country | Vol. | % | Country | Vol. | % | Country | Vol. | % |
| Mexico | 73,583 | 64.8 | Mexico | 59,937 | 36.7 | Mexico | 72,018 | 32.9 | Mexico | 75,016 | 37.2 | Mexico | 76,062 | 34.6 |
| Panama | 8,423 | 7.4 | India | 14,276 | 8.7 | India | 33,570 | 15.3 | India | 29,637 | 14.7 | Ecuador | 20,195 | 9.2 |
| El Salvador | 6,697 | 5.9 | Venez. | 12,719 | 7.8 | Panama | 11,613 | 5.3 | Panama | 9,787 | 4.8 | Panama | 13,727 | 6.2 |
| Ecuador | 4,192 | 3.6 | Panama | 10,264 | 6.2 | Venez. | 11,563 | 5.2 | Ecuador | 8,058 | 3.9 | India | 12,999 | 5.9 |
| Guyana | 3,568 | 3.1 | Guyana | 7,972 | 4.8 | Guyana | 10,165 | 4.6 | El Salvador | 6,787 | 3.3 | Thailand | 8,841 | 4.0 |
| Japan | 2,947 | 2.6 | Iran | 6,801 | 4.1 | Pakistan | 7,125 | 3.2 | Nicaragua | 6,178 | 3.0 | Brazil | 8,768 | 3.9 |
| India | 2,892 | 2.5 | Pakistan | 6,566 | 4.0 | El Salvador | 6,354 | 2.9 | Colombia | 5,712 | 2.8 | El Salvador | 6,233 | 2.8 |
| Colombia | 2,174 | 1.9 | Ecuador | 5,667 | 3.4 | Nicaragua | 6,021 | 2.8 | Guyana | 5,382 | 2.6 | Nicaragua | 5,624 | 2.5 |
| Iran | 1,226 | 1.1 | El Salvador | 5,376 | 3.2 | Ecuador | 5,992 | 2.7 | Venez. | 4,913 | 2.4 | Taiwan | 5,427 | 2.4 |
| Pakistan | 1,018 | .9 | French Guiana | 3,960 | 2.4 | French Guiana | 5,054 | 2.3 | Guatemala | 3,642 | 1.8 | Guyana | 5,281 | 2.3 |
| TOTAL | 106,720 | 93.8 | TOTAL | 133,538 | 81.3 | TOTAL | 169,475 | 77.2 | TOTAL | 155,112 | 76.5 | TOTAL | 163,157 | 73.8 |
| Grand TOTAL | 113,418 | | Grand TOTAL | 162,942 | | Grand TOTAL | 218,715 | | Grand TOTAL | 201,457 | | Grand TOTAL | 219,308 | |

[a] The percentage figures represent the percentage of total U.S. Shrimp imports supplied by the indicated countries.

increased Ecuadorian shrimp exports. Over 50 percent of the Ecuadorian production comes from shrimp farms (Graham, Empacadora Nacional, Ecuador, personal communication). Ecuador's exports to the U.S. have been steadily increasing from 5.7 million pounds in 1964 to 24.7 million pounds in 1981 (11.1 percent of U.S. imports in 1981).

Panama has consistently been a major exporter to the U.S. Between 1964 and 1979, Panamanian imports ranged between 10 and 12 million pounds annually but increased to nearly 16 million in 1981 (7). The U.S. has been, for many years, the principal market for Panama's shrimp exports, and it is expected to continue to be. Between 80 and 90 percent of Panama's production is exported, and 95 percent of that comes to the U.S. (2).

In recent years, Brazil and Thailand have become major exporters to the U.S., while Pakistan and Guyana have declined in importance. Overall, U.S. shrimp imports sources have become more diversified. In 1960, 93.8 percent of U.S. imports came from her top ten suppliers. By 1980, this share declined to 73.8 percent.

## U.S. IMPORT DEMAND AND FOREIGN SUPPLY

The import demand for shrimp in the U.S. determines the quantities that will be imported at various prices as world shrimp supplies change. A simultaneous estimate of U.S. import demand and world supply to the U.S. was made to analyze the effect of changes in relevant economic variables on the demand for and supply of shrimp entering the U.S. The demand, supply and equilibrium equations estimated for the 1960-81 period are as follows:

$$Q_d = \underset{}{.133}\ P^{\underset{(.125)}{-.290}}\ M^{\underset{(.482)}{2.037}}\ S^{\underset{(.172)}{-.114}}\ I^{\underset{(.080)}{-.180}} \qquad (1)$$

$$Q_s = 2.57(10)^5\ F^{\underset{(.205)}{.955}}\ J^{\underset{(.186)}{-.403}} \qquad (2)$$

$$P = FE \qquad (3)$$

$$Q_d = Q_s \qquad (4)$$

where:

Q = 1,000 pounds of imported shrimp (heads off)

P = U.S. dollars per pound of imported shrimp

M = U.S. real disposable income (100 million)

S = U.S. shrimp landings (1,000 pounds - heads off)

I = Beginning U.S. inventories (1,000 pounds - heads off)

F = Import shrimp price expressed in a weighted foreign currency

E = Reciprocal of the foreign exchange rate (weighted foreign currency per U.S. dollar of major exporters)

J = Japan import price expressed in a weighted foreign currency of principal exporting countries

The demand price coefficient, -.290, indicates the U.S. import demand is highly inelastic. If import prices are increased by 10 percent, the expected immediate effect would be a reduction in quantity imported of only 2.9 percent. Conversely, if the quantities exported to the U.S. are restricted either by export restrictions placed by foreign countries or import tariffs or quotas placed by the U.S. government, the price of imports will increase significantly. For example, a one percent reduction in quantity imported will result in a U.S. import price increase of approximately 3 percent.

U.S. demand for shrimp imports is highly responsive to consumer incomes. The estimated income elasticity, 2.037, indicates that a one percent increase (decrease) in real disposable income will result in an increase (decrease) of 2.037 percent in demand at existing prices and constant values for other demand variables.

The demand for imports is a derived demand in that imports enter into the marketing and processing sector for processing and/or sale through restaurants and other retail outlets. Domestic shrimp production and inventories are a substitute for imported shrimp. The coefficients for U.S. landings and beginning inventories in equation (1) indicate these substitutes have a negative effect on demand for imports. Beginning inventories have a significant effect on import demand: a one percent increase (decrease) in quantities held in beginning inventories will reduce (increase) import demand in that year by .180 percent. The negative effect of domestic landings on import shrimp demand is, however, not highly statistically significant. This may be because at least some of the domestic landings go into inventories. The impact of domestic landings then may be partially reflected by the estimated impact of beginning inventories.

Quantity supplied to the U.S. by foreign supplies is a statistically significant function of prices for imported shrimp, expressed in weighted average currency of principal suppliers (equation 3). The estimated coefficients indicate that a one percent increase (decrease) in foreign import prices will cause shrimp exporters to increase (decrease) the quantity supplied to the U.S. by .955 percent. In the present model formulation (equations 2 and 3), either a one percent increase (decrease) in the exchange rate (foreign currency per U.S. dollar) or U.S. dollar prices will increase (decrease) supply to the U.S. by .955 percent.

Shrimp import prices in Japan were included in the U.S. import

supply equation because Japan has become a major competitor of the U.S. for shrimp imports. Japan's shrimp imports increased from approximately one half million pounds in 1959 to 357 million pounds in 1981 (8). Prices paid by the Japanese therefore should affect the quantities suppliers are willing to supply to the U.S. at given exchange rates and U.S. prices. The estimated Japanese price effect in equation (2) has the expected negative sign and is statistically significant. The coefficient suggests a one percent change in Japanese import prices (in units of foreign currency) will cause a .403 percent change of opposite direction in quantity supplied to the U.S.

Equation (4) is the market clearing condition which requires quantity demanded to equal quantity supplied. It is required for the simultaneous estimation of demand and supply.

## IMPLICATION FOR THE U.S. SHRIMP MARKET

U.S. shrimp production has generally stabilized. Any substantial growth in U.S. consumption will have to come from increased imports in the near future. In recent years, Mexico and India have supplied approximately 40 percent of U.S. shrimp imports. These two countries appear to have reached maximum levels of shrimp output. Ecuador, Panama, Thailand and Brazil supply approximately 24 percent of U.S. import needs. These countries are experiencing growth in shrimp production. It is probable that some of their growth is from cultured shrimp production. Data are not available on a continuous basis for cultured shrimp. Ecuador seems to be the most successful country in production of cultured shrimp and is presently trying to expand acreage used in shrimp farming (H. Graham, Empacadora Nacional, Ecuador, personal communication). It thus appears that future growth in volume available for imports will depend on developments in shrimp culture and a continued trend by the U.S. in developing a more diversified supply of imported shrimp.

The U.S. demand characteristics for imported shrimp indicate substantial increases of import shrimp prices unless new abundant sources of supply materialize. The price parameter suggests prices will increase significantly compared to small reductions in quantity. Future growth in personal disposable income will increase the demand still farther for imported shrimp. Increases in demand for imports by countries such as Japan and the EEC will reduce the import supply offered to U.S. consumers.

Without substantial increases in world cultural shrimp production and/or discovery of new shrimp beds, consumers can expect to pay higher prices for shrimp. Evaluation of demand parameters tend to support these conclusions. The processing sector will likely be able to maintain present production levels due to the inelastic nature of import demand for shrimp. Growth in the processing sector is not probable given relatively stable supplies, the current high dependency on import shrimp for processing, and the current excess capacity in the processing sector.

To draw implications of the impact of imports on the harvesting sector, one final model needs to be developed. A double log model expressing exvessel prices (XP) received by fishermen in the Gulf and South Atlantic was estimated to be a function of consumer disposable incomes (M), Gulf and South Atlantic landings (G), beginning inventories (I), and volume of imports (V). The model expressed as equation (4) explained 93 percent of the variation in annual exvessel prices for the 1960-81 period. Estimated parameters and standard errors are:

$$XP = (2.96)(10)^{-5}\ M^{4.791}\ G^{-.664}\ I^{-.549}\ V^{-1.398} \qquad (4)$$
$$(.517)\quad (.221)\quad (.137)\quad (.382)$$

The three supply variables; G, I and V; all have significant negative impacts on prices fishermen receive. As these supply sources become more stable, these factors will tend to have less downward pressure on shrimp prices. There is a strong relationship between quantity of imports and exvessel prices; a 10 percent reduction in quantity imported would increase exvessel prices by nearly 14 percent. Future growth in consumers' real disposable incomes will tend to increase prices received by fishermen. An increase of one percent in consumers' real incomes after taxes will result in an increase of 4.791 percent in exvessel prices.

## REFERENCES


1. ANONYMOUS, Aug. 1982. India: an aggressive attitude. The fish boat. Harry L. Peace, publisher. Vol. 27(8). Covington, Louisiana.

2. DIRECCION GENERAL DE RECURSOS MARINOS, 1979. Estadistica pesquera 1969 a 1978. Ministerio de Comercio e Industrias. Republica de Panama.

3. FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS (FAO). 1950-80. Yearbook of fishery statistics. FAO, U.N., Rome.

4. GULLAND, J.S., 1971. The fish resources of the ocean. Food and Agriculture Organization of the United Nations. Fishing News (books) Ltd., publisher. Surrey, England, p. 240.

5. NATIONAL MARINE FISHERIES SERVICE, June 1982. Shellfish market review. U.S. Department of Commerce. U.S. Government Printing Office. Washington, D.C., 47 pp.

6. NATIONAL MARINE FISHERIES SERVICE, 1964-81. Imports and Exports of fishery products. U.S. Department of Commerce. U.S. Government Printing Office. Washington, D.C.

7. NATIONAL MARINE FISHERIES SERVICE, 1960-81. Fisheries of the United States. U.S. Department of Commerce. U.S. Government Printing Office. Washington, D.C.

8. STATISTICS AND INFORMATION DEPARTMENT, 1965-81. Fisheries statistics of Japan. Ministry of Agriculture, Forestry and Fisheries. Government of Japan. Tokyo.

# WORLD PRODUCTION, IMPORTS AND U.S. DEMAND FOR SPINY LOBSTERS

Fred J. Prochaska and Walter Keithly
Department of Food and Resource Economics
University of Florida

## INTRODUCTION

Spiny lobsters are one of the most valued seafood commodities in the U.S. Among the major species landed in the U.S., spiny lobsters trailed only bay and sea scallops in exvessel prices in 1981. Among imports in the fresh and frozen categories of fishery products, spiny lobster was easily the highest valued product on a per pound basis. Lobster imports, valued at $225 million, were exceeded in total value by only imports of total groundfish, blocks and slabs, tuna and shrimp (6).

The market for domestically landed spiny lobster is expected to be significantly influenced by the import market because over 90 percent of the spiny lobster consumed in the U.S. is imported. Florida is the leading domestic producer of spiny lobsters, with California the only other regular producer of any significance (2).

The purposes of this paper are to (1) review total world production and U.S. imports of spiny lobster, (2) analyze U.S. demand for spiny lobsters, and (3) determine the impact of the U.S. spiny lobster market on exvessel demand for spiny lobsters in the domestic fishery. For purposes of this study, all world production and imports of lobsters other than Homarus lobsters are considered to be spiny lobsters.

## U.S. SPINY LOBSTER IMPORTS

Total imports of fresh and frozen spiny lobsters increased from slightly over 32 million pounds in 1960 to 48.5 million pounds (product weight) in 1976 (4). After 1976, total imports began to decline through 1980 when a total of only 36 million pounds were imported.

Total fresh and frozen tail imports increased from 27.9 million pounds in 1960 to a high of 37.3 million pounds in 1969. Since 1969, total tail imports have trended downward, reaching a low of 27.4 million pounds in 1980. Because of an increase in volume of miscellaneous spiny lobster products, tail imports dropped from approximately 87 percent of total imports in the early 1960s to an average of 73 percent since 1976.

Spiny lobster imports are further classified into cold water imports and warm water imports. Differences between these two classes of lobster imports further explain trends in the total lobster import market.

## Cold Water Imports

Cold water imports increased from approximately 20 million pounds in 1960 and 1961 to a record of 28.1 million pounds in 1968 (Figure 1). Between 1968 and 1970, cold water imports declined sharply to 19.3 million pounds. After 1970, cold water imports declined at an annual average rate of approximately one third million pounds per year and reached a low of 15.5 million pounds in 1980.

Cold water imports accounted for 62 percent of total U.S. imports during 1960-61 and reached a high of 67 percent in 1967. By the 1979-80 period, cold water imports declined to 41 percent of the total. Essentially all cold water imports are in tail product form. In 1967, miscellaneous cold water products were at a maximum of 2.9 million pounds (Figure 1). Since 1970, cold water miscellaneous products have generally been less than 1.0 million pounds annually.

Cold water lobsters come from Australia, South Africa and New Zealand (Figure 1). South Africa was the leading cold water lobster exporter during the 1960s and accounted for nearly one half of total cold water imports to the U.S. Between 1968 and 1972, South African cold water imports to the U.S. declined 60 percent to 4.5 million pounds, due in part to restrictions placed on the fishery (5, June 1970). After a small recovery, South African imports declined to a low of 2.8 million pounds in 1980.

Australia has been the leading cold water lobster exporter to the U.S. since 1970 (Figure 1). Between 1960 and 1980, Australian imports increased from approximately 8 million pounds to 10 million pounds. New Zealand imports increased through 1969 but then declined to levels equal to the early 1960s by the latter part of the 1970s.

The peak in total cold water imports in 1968 came about because of increased imports from all three countries. Since 1968, the total decline has come from reduced imports from South Africa and New Zealand. With the apparent stabilizing of Australian and New Zealand imports, any further decline in total cold water imports will likely come from further reductions in South African imports. However, since South African imports are only approximately 3 million pounds currently, any decline will be minor.

## Warm Water Imports

Warm water lobster imports are exported to the U.S. by anywhere from 20 to 30 countries in recent years. Many of the major exporters are Latin American countries, with each country shipping

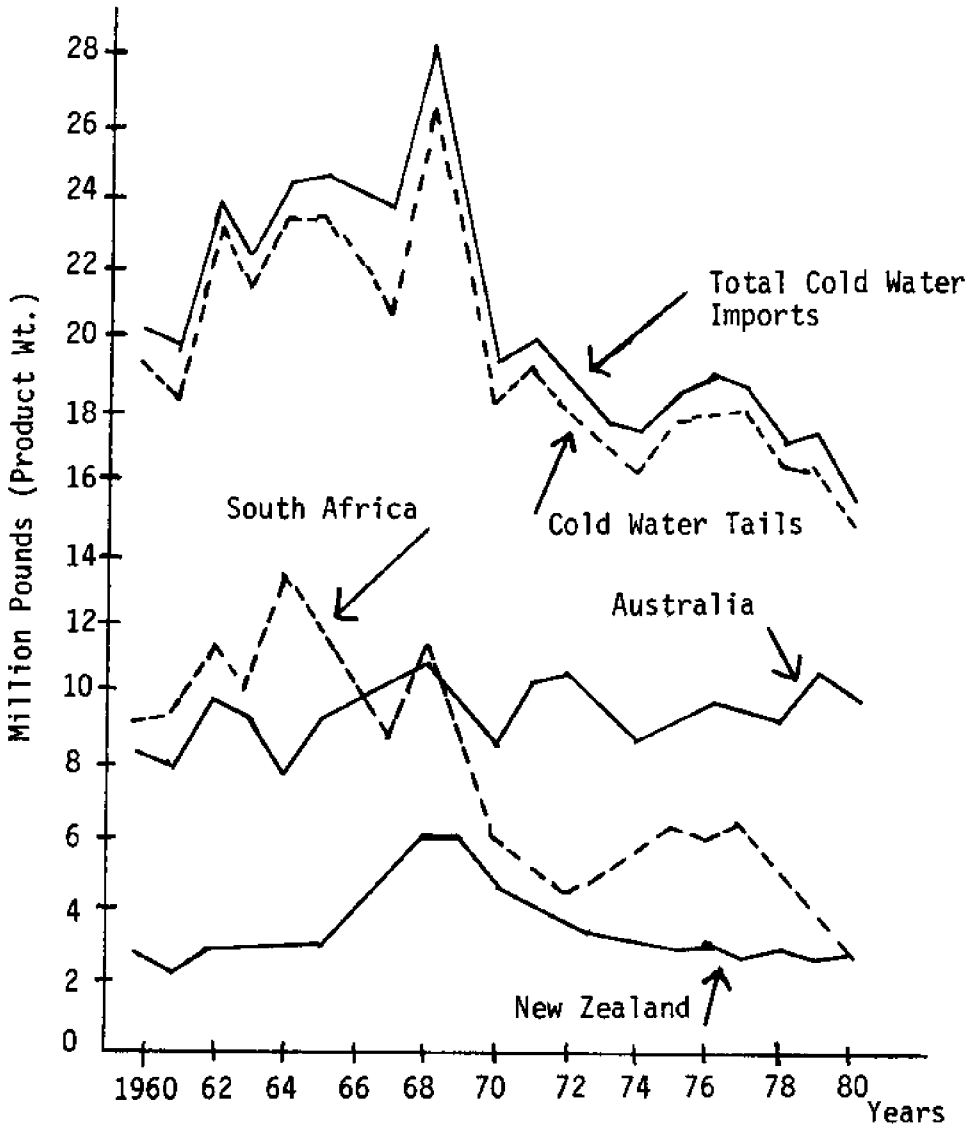


Figure 1. U.S. Cold water lobster imports by country, 1960-80. (Derived from 4).

relatively small quantities. Total warm water imports into the U.S. increased significantly over the past two decades. Imports ranged from 10 to 12 million pounds annually between 1960 and 1964 (Figure 2). After 1964, total warm water imports increased rapidly to a maximum of 29.4 million pounds in 1976. Imports then declined to around 26 million pounds for the next three years but then fell sharply in the last year to 20.5 million pounds.

Warm water tail imports gradually declined between 1960 and 1967 and generally accounted for 50 to 80 percent of total warm water imports (Figure 2). After 1967, warm water tail imports increased rapidly to over 14 million pounds in 1969 but since then have remained relatively stabile with annual imports generally ranging from 14 to 16 million pounds. Miscellaneous warm water imports increased from 2.8 million pounds in 1960 to a high of 14.0 million pounds in 1976. Imports then appeared to stabilize around 10 to 12 million pounds but then declined significantly in 1980.

Brazil has been the leading exporter of warm water lobsters to the U.S. throughout the 1960-80 period. Brazil's exports to the U.S. averaged around 3.3 million pounds during the 1960s but then increased rapidly in 1969. After 1969, her exports generally were between 5 to 7 million pounds annually. Mexico and Chile are the only other exporters of warm water lobsters to the U.S., with a consistent volume over 1.0 million pounds annually. Mexican imports to the U.S. gradually increased from 1.6 million pounds in the early 1960s to approximately 2.1 million pounds currently. In recent years, approximately two thirds of Mexican imports and almost all of Chile's imports are classified as miscellaneous products, while nearly all of Brazil's exports to the U.S. are tails. Since 1975, Honduras and Nicaragua each have exported over 1.0 million pounds to the U.S. annually. Exports from Honduras reached a high of 2.6 million pounds in 1979, while exports from Nicaragua reached a high of 2.8 million in 1978. Countries supplying the remaining 10 to 12 million pounds of warm water imports into the U.S. in recent years include Latin American, South American, European and Asian countries and some islands.

## PRICES AND VALUE OF LOBSTER IMPORTS

Prices of imported spiny lobsters and the total value of imports have increased considerably during the past two decades. The average import price of cold and warm water lobster imports increased from $ .96 per pound (product weight) in 1960 to $6.43 per pound in 1980 (Figure 3). Cold water prices have always been above warm water lobster prices throughout the time period considered. Warm water lobster prices were 27 cents per pound below cold water prices in 1960.

Both warm water and cold water prices increased gradually through 1970. After 1970, prices increased at an annual average rate of 46 cents and 28 cents per pound for cold water and warm water lobsters, respectively. Prior to 1970, import prices increased at a rate slightly above the consumer price index (CPI).

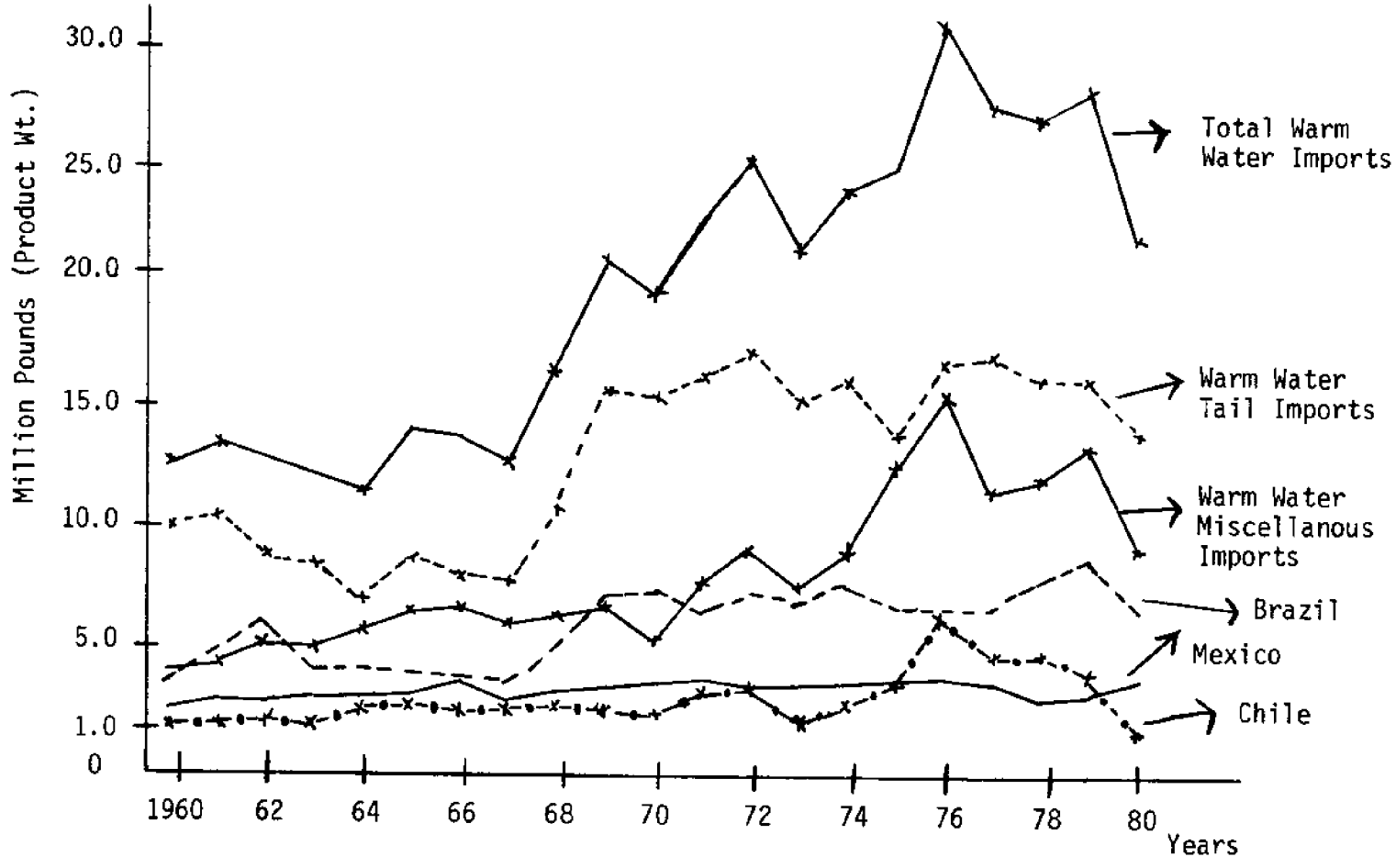


Figure 2. U.S. Warm water lobster imports by country. (Derived from 4).

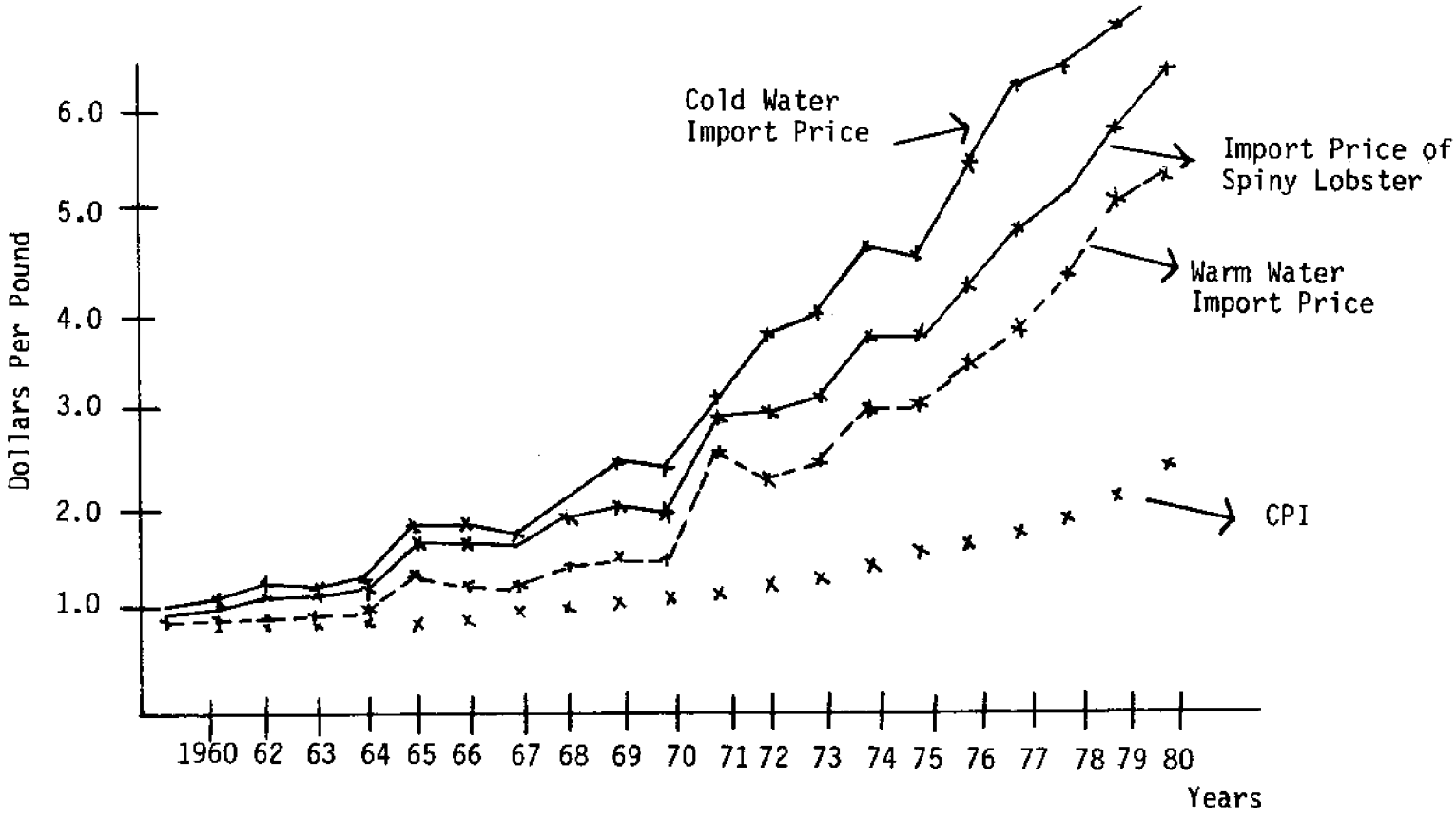


Figure 3. Spiny lobster import price in relation to the consumer price index. (Derived from 4).

After 1970, the rate of increase in lobster prices has been 3.2 times the rate of increase in the CPI.

Total value of spiny lobster imports increased from $30 million in 1960 to an average of approximately $240 million in 1979 and 1980 (Figure 4). From 1960 through 1967, the increase in total value of imports was due to increases in volume imported and moderate increases in prices but, since 1968, essentially all of the increased value has been due to price increases. After inflation has been accounted for, the real value of total spiny lobster imports increased to an average of $120 million annually during the late 1970s. This represents an increase in real value of 300 percent over the 10-year period.

Price increases for cold water tails have more than offset the decline in cold water import value. The value of cold water lobster imports increased from a little over $20 million in 1960 to slightly over $120 million in 1980. Value of warm water imports were generally less than one half the value of cold water imports during the 1960s; however, by the second half of the 1970s, the average annual values were equal.

## DOMESTIC PRODUCTION

Florida accounts for essentially all of the U.S. domestic production of spiny lobsters (2). The major species landed in Florida, the Panulirus argus, is considered a warm water species. Landings increased from 2.9 million pounds in 1960 to 11.4 million pounds in 1972 (Figure 5); a gain of 282 percent. In 1975, however, landings of spiny lobster in Florida fell sharply and since then have averaged about 6.5 million pounds, compared to the 10.3 million pounds for the five years preceeding 1975 (Figure 5). The decline came about for two reasons (7). First, the Bahamian waters were closed to Florida fishermen as a result of the 1974 Bahamas Fisheries Amendment Act. Second, domestic commercial landings of spiny lobster were a maximum and have since then remained relatively stabile or declined. Given the status of the Bahamian fishery and domestic fishery, increases in domestic landings are not expected.

Value of Florida landings increased from approximately $1.0 million in 1960 to $15 million in 1979 (Figure 5). Annual movements in value generally followed movements in landings through the early 1970s. Since that time, value of landings has increased even though volume of landings has declined. Florida exvessel prices increased over 435 percent from $ .39 per pound (whole weight) in 1960 to $2.10 per pound in 1980.

## DOMESTIC DEMAND

U.S. demand for spiny lobsters is unique in that spiny lobsters are one of the few food products that world production is mainly consumed by one country. In this section, U.S. production and imports are first considered with respect to total world production. Economic determinants of U.S. demand are then analyzed.

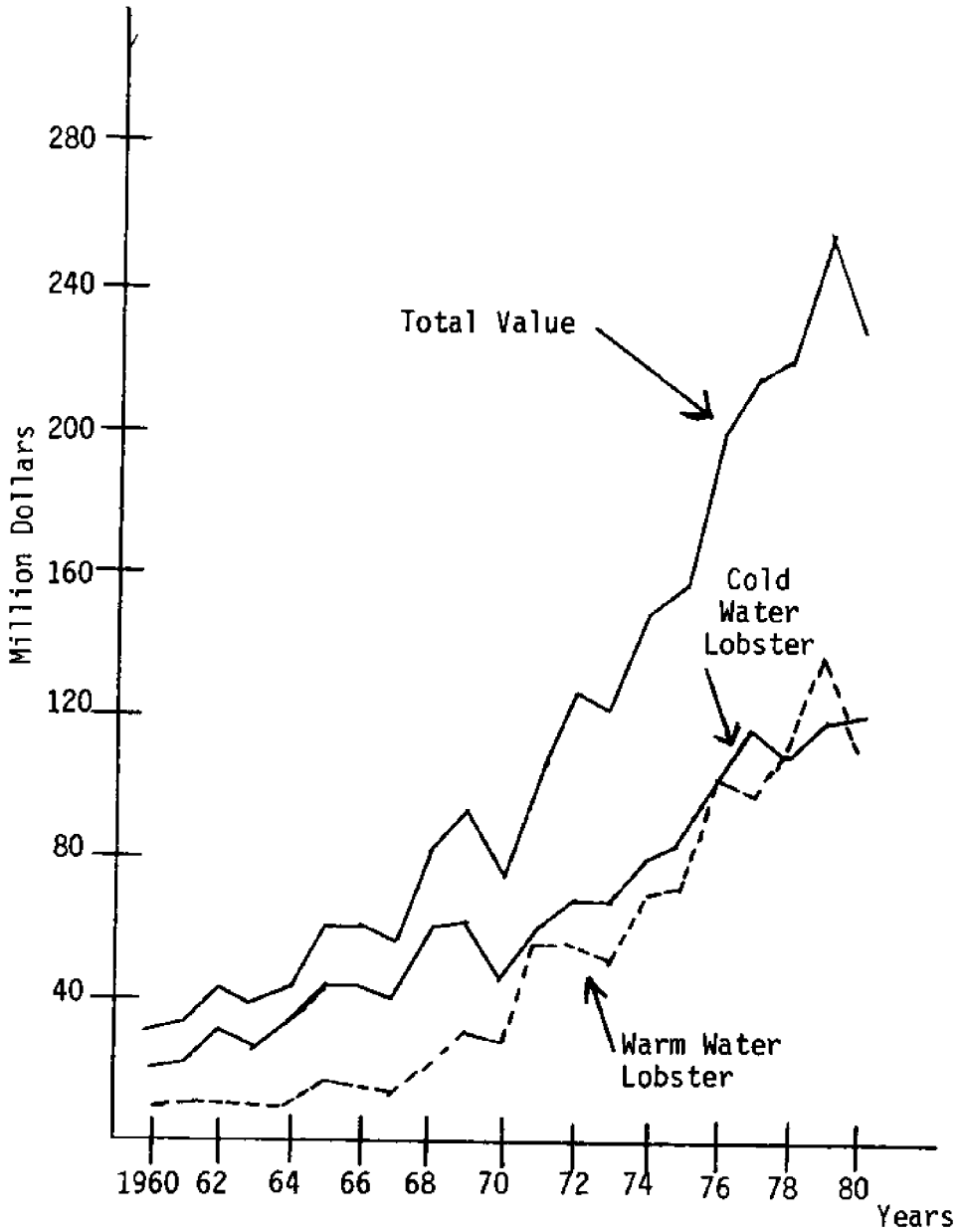


Figure 4. U.S. Value of spiny lobster imports, 1960-1980. (Derived from 4).

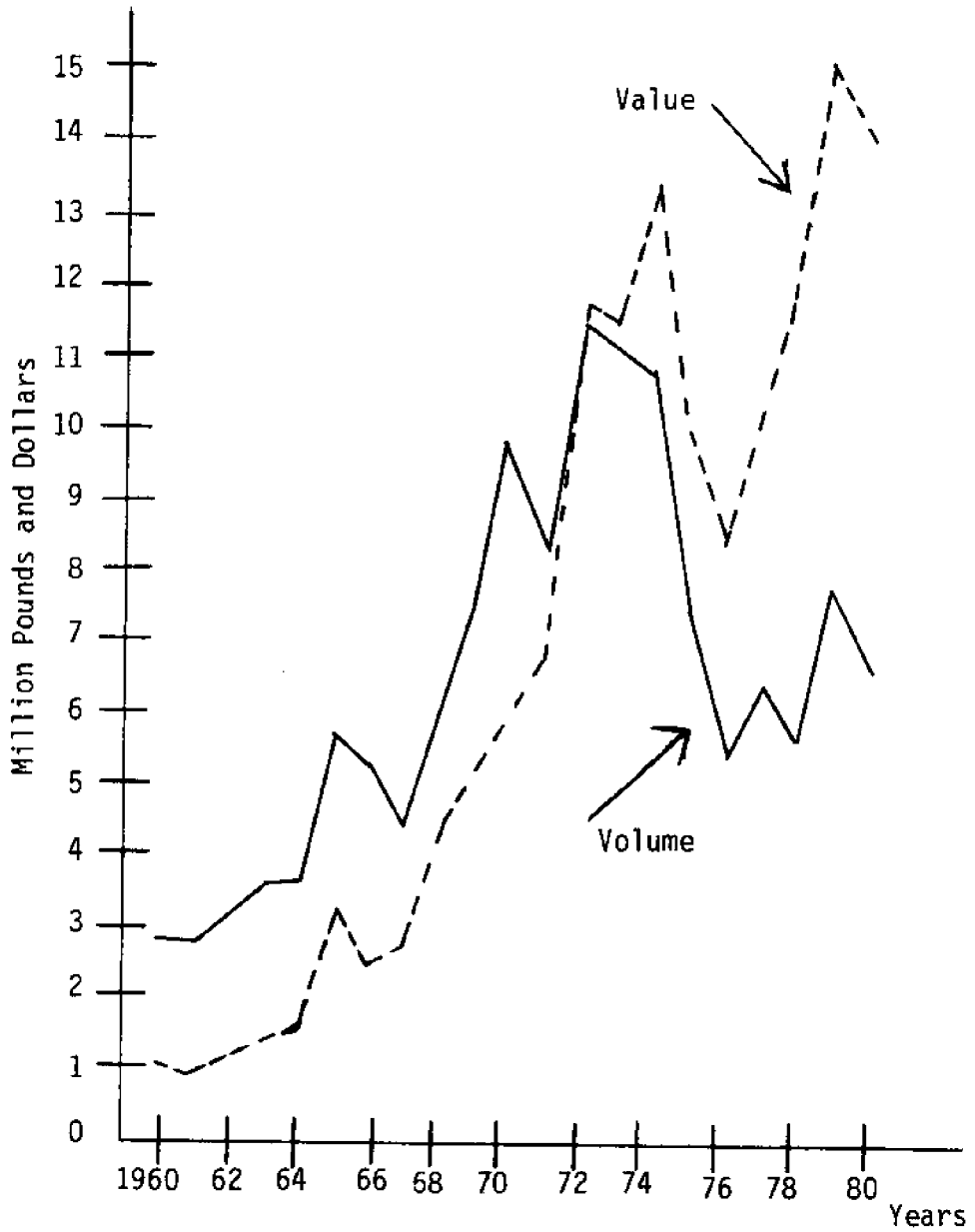


Figure 5. Florida spiny lobster landings and value, 1960-1980. (Derived from 2).

## World Production

Total world production of spiny lobster expressed in tail weight[1] trended upward over the past two decades from approximately 38 million pounds to 50 million pounds (Figure 6). During the 1960s, increases in total production were a result of increased landings of cold water lobsters. After 1970, increased landings of warm water lobster more than offset declining cold water landings, which allowed total world production to increase.

U.S. imports of spiny lobsters and U.S. production jointly account for most of the world production. During the 1960 through 1979 period, imports plus domestic production accounted for 82 percent or more of total world production. In the peak import years of 1959 and 1976, the data suggest that imports plus U.S. production equaled or exceeded world production. For these years, there could be errors in the data or imports could have been from carry over of production from the previous year. Comparison of U.S. annual imports with production trends in cold or warm water lobsters indicates a lag response of imports to production in some years where production increases were substantial. One further possible explanation for U.S. supply exceeding estimated world production in these years is the assumption that all world production was exported in tail weight equivalents. If in those years substantial imports of whole lobsters occurred, the discrepancy could be explained. U.S. imports relative to total world production was approximately 74 percent in 1980 and was the minimum share of world production accounted for by the U.S. The Japanese were reported to have increased lobster imports substantially in 1980 (5, September, 1981).

U.S. cold water lobster imports (Figure 1) account for essentially all cold water lobster production (Figure 6). The long-term trend and annual variations in cold water imports generally parallel production. U.S. imports of warm water tails (Figure 2) were equal to approximately 55 percent of world production (Figure 6) during the early 1960s. By the 1976-79 period, the U.S. imports accounted for an average of 93 percent of total world production of warm water lobsters.

## Price and Income

Theoretically, for given tastes and preferences, prices and per capita disposable income are the principal factors affecting demand for a product. The demand for lobsters in the U.S. was estimated to be a function of these variables in the following equation:

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[1] Tail weight was estimated as one third the volume expressed in round weight.

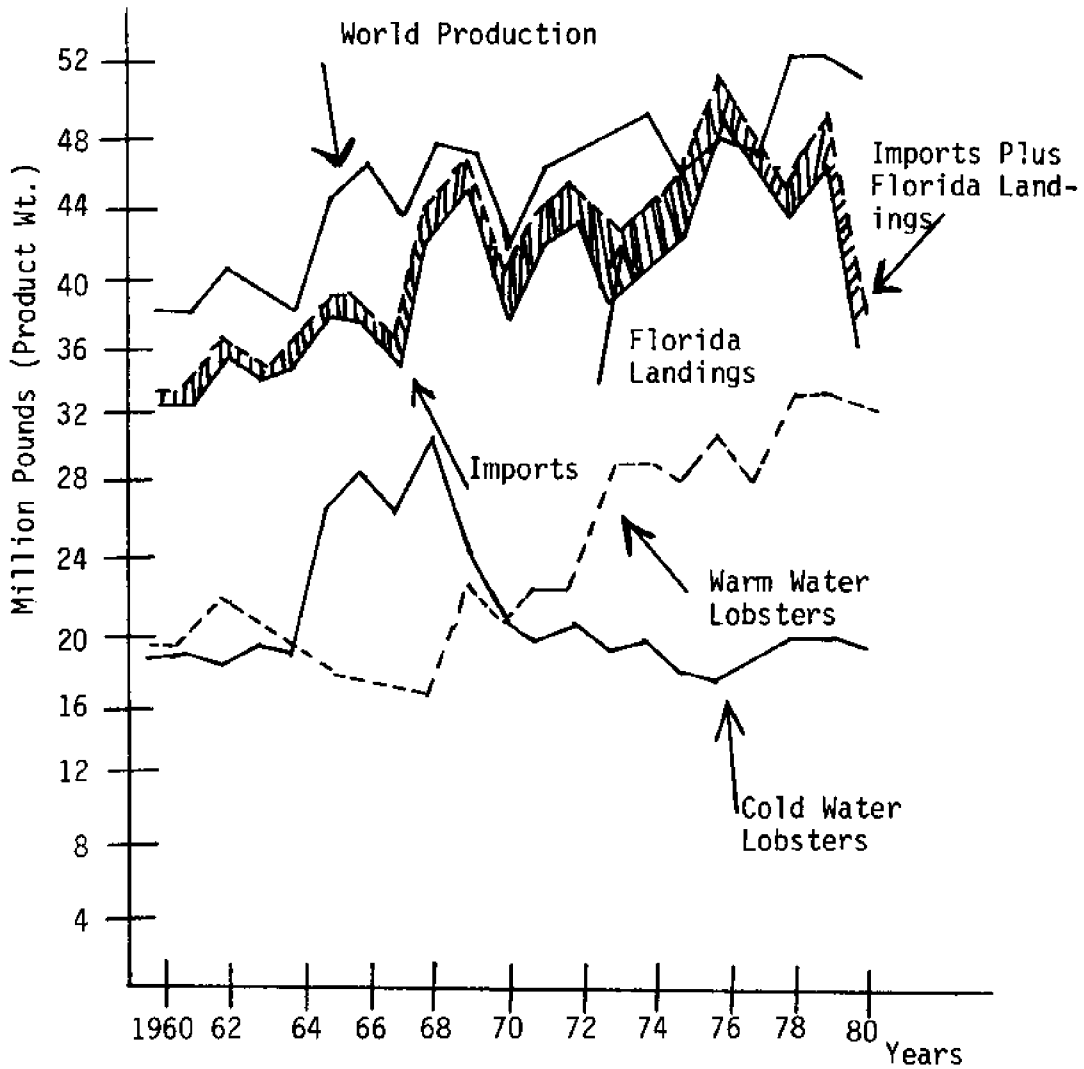



Figure 6. Spiny lobsters production and U.S. imports 1960-80. (Derived from 1, 2, and 4).

$$Q = 845.56M^{\underset{(.665)}{1.371}} P^{\underset{(.403)}{-.696}} \quad (1)$$

where:

Q = million pounds of lobsters consumed annually,

M = per capita disposable (after tax) real income measured in $1,000 units, and

P = weighted average lobster prices in dollars per pound.

Quantity consumed each year, Q, was estimated as the sum of spiny lobster and American lobster produced and imported plus beginning inventories minus ending inventories. Income was deflated by the consumer price index (1973 = 100). Annual average prices were import prices of each type of lobster weighted by quantity of American lobster and quantity of spiny lobster (both domestically produced and imported). Import prices were used because of the dominance of imports in the spiny lobster market and because import prices are more closely related to consumer prices than are exvessel prices. Consistent series of retail prices do not exist. Import prices were used for American lobsters in order that all prices would be at the same market level. Data used covered the 1961-1980 time period. The multiple correlation coefficient was .27.

The price elasticity of -.696 in equation (1) is statistically significant at the 95 percent confidence level and is consistent with previous estimates presented in a 1974 publication (3). The demand is inelastic with respect to price. A one percent increase (decrease) in price will result in a decrease (increase) in consumption of .696 percent. This demand characteristic indicates price will vary widely for relatively small changes in total supply.

The relatively large elastic demand for lobsters with respect to income suggests lobster consumption is responsive to changes in real income. The income elasticity of 1.371 indicates that, at given prices, a one percent increase (decrease) in real per capita disposable income will result in an increase (decrease) of 1.371 percent in lobster consumption. The estimated income elasticity was statistically significant at the 90 percent confidence level. The income elasticity estimate is less than the 1974 estimate which was 1.95. That estimate was at the exvessel level of prices which are farther removed from the consumer market level than the wholesale prices used in the present analysis. Furthermore, in the earlier estimate, the data covered a time period when miscellaneous products were of less importance in total lobster consumption. These differences probably explain the difference in size of the estimated income elasticities.

Numerous attempts, including simultaneous supply and demand models, were made to analyze the wholesale or import demand for

spiny lobsters separately from American lobsters without success. Separate estimates were attempted for total spiny lobster imports, cold water imports and warm water imports. The individual estimates generally resulted in significant positive income elasticities but generally had insignificant negative price elasticities. Prices of the different species, being highly correlated, may have led to this inconsistency (2).

The demand for spiny lobster at the exvessel market level was estimated as a price dependent model. Estimated parameters and standard errors are presented in equation (2).

$$EP = .255PI^{\underset{(.046)}{1.069}}\ QL^{\underset{(.062)}{-.123}} \qquad (2)$$

where:

EP = exvessel prices in dollars per pound,

PI = price of imported spiny lobsters in dollars per pound, and

QL = annual quantity landed in Florida.

The model explains 98 percent of the variation in annual exvessel prices for the 1960-80 time period. The crossprice flexibility, 1.069, suggests nearly a one-to-one movement between import prices and Florida exvessel prices. The flexibility, 1.069, indicates a one percent increase (decrease) in import prices will increase (decrease) exvessel prices by 1.069 percent. The quantity landed variable indicates exvessel prices will decrease (increase) .123 percent for a one percent increase (decrease) in landings. Income was not included in the equation to explain exvessel prices because income, at least in part, determines import prices which are included in model (2). This specification assumes income more directly affects import prices than exvessel prices. This seems reasonable since imports make up over 90 percent of spiny lobster consumption in the U.S. These conclusions strongly suggest that prices received by domestic spiny lobster fishermen are highly dependent on the import market. Although the quantity landed had a statistically significant effect on exvessel prices, the economic size of the effect is minor.

## SUMMARY AND CONCLUSIONS

World production of spiny lobsters appears to be reaching a maximum. Cold water spiny lobster landings have declined during the past decade while warm water landings appear to have stabilized after 1973. Until 1980, the U.S. imported most of the world production. In recent years, there appears to be increased competition for world supply; principally by Japan. These events have resulted in decreased imports to the U.S. after 1976. Reduced imports and stabile domestic production suggest a decline in U.S. supply of spiny lobsters or, at best, a stabile supply.

Demand estimates indicate prices will increase substantially if supplies are reduced. Reduced supplies may come about from overfishing of world resources and/or increased import competition from other countries. Increased real incomes in the U.S. will further increase the demand for lobsters. With stabile supplies, prices will be increased further due to the income effect. Increased prices will be transmitted to the dockside level at a rate faster than the general rise in consumer prices if present trends continue since exvessel prices move nearly in a one-to-one ratio with import prices. Any actions by domestic producers to affect prices through controls on domestic production will have minimal effects.

## REFERENCES


1. FOOD AND AGRICULTURAL ORGANIZATION, 1960-1980. Yearbook of fishery statistics: catches and landings. Food and Agriculture Organization of the United Nations. Rome, Italy.

2. GULF OF MEXICO AND SOUTH ATLANTIC FISHERY MANAGEMENT COUNCILS, 1981. Fishery management plan environmental impact statement and regulatory impact review for spiny lobster in the Gulf of Mexico and South Atlantic. Gulf of Mexico Fishery Management Council. Tampa, Florida.

3. NATIONAL MARINE FISHERIES SERVICE, 1974. American and spiny lobsters, 1947-73: basic economic indicators. National Marine Fisheries Service, NOAA, Curr. Fish. Stat. 6272. Washington, D.C.

4. U.S. BUREAU OF CENSUS, 1960-1980. U.S. imports for consumption. U.S. Department of Commerce, Bureau of Census. U.S. Government Printing Office. Washington, D.C.

5. NATIONAL MARINE FISHERIES SERVICE (Various Issues). Shellfish market review. National Marine Fisheries Service, NOAA, Curr. Econ. Analysis. U.S. Government Printing Office. Washington, D.C.

6. NATIONAL MARINE FISHERIES SERVICE, 1981. Fisheries of the United States, 1981. National Marine Fisheries Service, NOAA, Curr. Fish. Stat. 8200. U.S. Government Printing Office. Washington, D.C.

7. PROCHASKA, F. J. and J. S. Williams, 1978. Economic analysis of spiny lobster firms at optimum stock levels. Southern Journal of Agricultural Economics 10(December): 93-100.

# WAYS OF IMPROVING COMMUNICATION (EXTENSION) BETWEEN THE FISHERIES INDUSTRY AND INSTITUTES IN LATIN AMERICA

Héctor M. Lupín
Fish Utilization and Marketing Service
Fisheries Department, FAO
00100 Rome, Italy

## INTRODUCTION

The application of basic knowledge to improve production is probably one of the weakest steps in the chain that forms the fishing activity in developing countries. The knowledge very often seems to be on hand but it rarely contributes to improving product quality or preventing loss or solving chronic export problems, etc., even though in many cases there are no economic reasons why the desired changes cannot be put into effect.

After years of multilateral and bilateral aid and its own efforts, nearly every Latin American country as well as many developing countries of Asia and Africa have teams of well trained people in all the disciplines of the fish industry. It is not rare to find people from developing countries with degrees from well-known American and European universities and, though not as frequent, research papers on fish fish handling, processing, etc., published in the world's top journals of food science and technology from countries such as Ghana, the Philippines and Peru, to mention only a few. Moreover, many countries in Latin America not only have research centres but also offer courses and careers at university level where the knowledge is stored, classified, refined and transmitted to others.

Nevertheless, the preparation and capacity of the people trained in these disciplines are not generally reflected in the fish industry and products of the country. More often, the following is the case.

Quite some time ago, I undertook a short, specific mission to a tropical country to supervise a programme being carried out by the natives but funded by an international organization. The Director of the local research institute greeted me with the following words: "Please, don't tell us we should use ice to keep the fish fresh, we already know it.". (The Director holds a Ph.D. from an American University.) I was somewhat surprised by his greeting since instructions on how to keep fish fresh was not the scope of my mission. So I asked him why he was telling

me this. He explained: "Because this is the first thing that international experts tell us upon arrival even though it may not have anything to do with their mission. We don't need this advice as we know all about icing fish, perhaps even better than the experts.". After finishing my work, I visited some landing places, fish markets and fish processing industries. Just before leaving I saw the Director of the institute again and told him that since I observed no one was using ice I was not surprised to hear that experts recommended it.

Although the above example can serve to analyse many different things, e.g. the role of internal and international communication, it also emphasizes the division that can frequently occur between research institutes or universities and industrial realty in developing countries. In this particular case, despite the Director's pride in his institute and the expertise of his staff, the fish industry was not only suffering from a severe loss of fish but also from a constant rejection of the country's exported fish products. It is doubtful that an increase in the support or staff of this institute and/or their technical training over a limited time span could improve the condition of their industry.

Of course, we must not put all the blame on the institutes. Many reasons which originate not only in the institutes but also in the industry and sometimes even in the governmental bodies dealing with fisheries help to sustain this situation. As a matter of fact, we could present a very different case and ask why an industry that is aware of its own problems does not ask for advice from the institutes that are right there or why fishery officers responsible for the government's fisheries policy do not encourage industry and institutes to work together.

## PROBLEMS THAT HINDER COMMUNICATION

The problem is the lack of proper communication between research institutes and industry. If this internal communication (extension) is not achieved, any self-sustained development will be very difficult no matter how much knowledge (external transference of technology) or inversions are poured in from foreign or national sources.

The first step for obtaining a good communication between institutes and industry in developing countries is to identify the problems that hinder it. These can stem from the institutional side, from the industrial side and from the governmental side. And we can include the common problems related to how people see technology.

### How People See Technology

We are living in a technological era at all levels and to all extents. What does this really mean in the everyday world? Basically, that everyone feels free to give his or her opinion on technology. We can compare it to the Renaissance era when

everyone felt free to voice their opinion on Leonardo's or Boticelli's paintings. Now everybody feels free to give their opinion on technology.

While people use caution before giving their opinions on medicine or law (in many countries they risk jail if they do), or people feel one should be a connoisseur before giving opinions on paintings and sculpture, they fell no such impediments to discuss technology, not matter at what level. Consequently, the political, social and practical implications are tremendous.

The technologist suffers when forced to present his proposals in plain language so they can be understood and discussed by managers, economists and people in general. If he fails to do this and tries to apply his knowledge directly he becomes a "technocrat". If he retreats and closes himself in an ivory tower he will be equally criticized but his life will be easier.

It is not a bad thing in itself that people are interested and feel free to discuss technology but limitations should be set and borne in mind in order to ensure a good communication. In developed countries this problem is overcome by the intermediate institutes between research and industry (e.g. extension services, R&D industry branch, consultants, etc.) but in developing countries technologists are directly faced with industry (7).

## Extension

Probably the first thing wrong with the word "extension" or its equivalent "transfer" is when it is used in "transfer of technology" (3). In order to transfer a thing, you must have a gradient. This means a difference between the person who "transfers" (the technologist/extensionist) and the person who receives (the producer). But do we always have a gradient when we try to introduce a technology?

Not really. The following physical analogy might help to clarify what is wrong with such a concept. The transference as stated above is like transferring water freely from one tank to another as shown in Fig. 1a. Nothing inside tank 2 can have an effect on the behaviour of tank 1.

But let us consider a different approach to the flux of technical knowledge. The first thing to realize is that the "extensionist" is at the same level as the producer. He will teach and he will learn about the subject. He will help and he will be helped in his/her work. Turning to another physical analogy, this is not the case of two separate tanks placed one over the other but of interacting tanks or communicating glasses (see Fig. 1b).

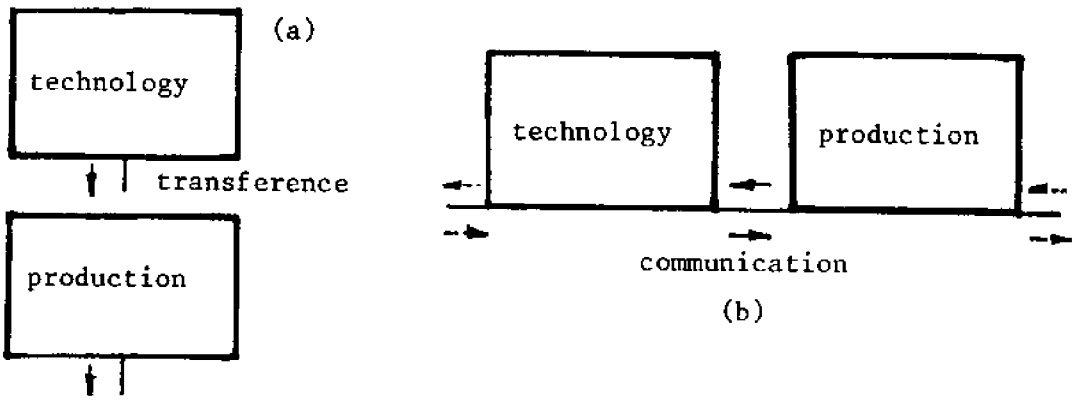



Fig. 1 Physical analogies to show a) the wrong way to see transference of technology and b) the interacting way.

Is there something the technologist/extensionist can learn from the producer? Definitely. The producer at industrial or artisanal level is continuously faced with the problems of production not only with regard to raw fish but also with economic constraints, cultural facts, etc. An extensionist will hardly have all this knowledge at his disposal.

This means that the "level" (knowledge) of the extensionist can be higher in certain subjects and the "level" (knowledge) of the producer higher in others. This interchange of knowledge, communication, when it exists, becomes a true means for improving not only production but also knowledge, if rightly applied.

People, intuitively or on purpose, usually reject the conventional concept of extension. The reaction varies widely, but we can identify two extremes. The first extreme relates to industrial production and the second to artisanal production.

Industrial production is normally carried out in developing countries (when national) as a mimetic of the same activity being carried out in developed countries (e.g. without innovations, with the result that production lines remain static for years). The owner, and sometimes the technicians involved, knows something about the basics of the process (e.g. canning) although the operations are mechanical and improvements rarely studied. With this superficial technological knowledge, the industrialist will usually refuse extension.

Who could possibly know more than he does about his country's tuna canning? "Surely, this extensionist is a newcomer who will try to steal my industrial secrets." This impression is usually quickly re-inforced as the extensionist can hardly be expected to possess the practical knowledge necessary for running an actual factory.

The industrialists tend to try to trap the extensionists by asking them, for example: "What is the best trade name of a pump to be used in a brine tank?" "What kind of oil should one use in the gear box of a sealing machine?" etc. The extensionist finds it very difficult to cope with these kind of questions, especially if they involve trade name comparisons and so on. So the industrialist becomes convinced that the extensionist doesn't know anything about the subject. The questions may have nothing at all to do with the useful knowledge the extensionist has to offer. Nevertheless, for the industrialist (or technical person involved) this is the best excuse for rejecting any idea he classifies as an invasion.

Certainly, extension in this classical sense is considered a cultural invasion. Unfortunately, this holds even truer for artisanal production (3) (9) (11).

At first glance, the situation of artisanal production often appears to differ from industrial. While they are attending training courses, people seem kind and cooperative, assist in meetings and demonstrations. And sometimes, someone even puts into practice the new techniques taught during the course (mainly if pushed). But enthusiasm soon disappears as soon as the extensionist goes back to the city. A common feature characteristic is that no one asks questions, they only make affirmations, e.g. "We will do it! No problem! etc., but at the same time no one is seriously thinking of how to accomplish the changes.

This is mainly why the word "communication" is replacing "extension". But more important than changing a word is to change attitude and approach when confronted with the problem of introducing new technologies.

## Language for Communication

Communication is an interchange of thoughts and opinions and language is the most important means of communication. But what should the level of effeciency of a language be?

The technologist and the producer (at industrial/artisanal level) should share the same vocabulary. Moreover, the same words should mean the same things otherwise communication will either not be possible or extremely limited, even if the same language is spoken.

The problem of language has been very well described when referring to extension at artisanal level (mainly in agriculture) (4) (5). But what I would like to point out here is that the problem also exists at industrial level in developing countries and even between different kinds of professionals dealing with the same subject.

Keeping language as simple as possible for communicating at artisanal level is a very good suggestion for any level. However,

a more elaborate approach is needed for coordinating communication between professionals in different fields and for communicating at industrial level.

The more complex the industry, the more complex the language. The technological level of the industry dictates the level of the language needed to establish communication. People in developed countries working in the fish industry and holding managerial positions are usually university graduates. But people in developing countries holding key posts in industry usually do not have university degrees and, hence, they cannot be expected to communicate with universities and research centres in a language that is academic or technical.

## The Definition of the Problem

Defining the problem is probably the first step to be considered by both the producers and extensionists when introducing a new technology. As in any other activity involving communication, how to begin is the most difficult part. Once this is overcome, the rest flows naturally. So the definition of the problem is decisive.

Even if problems of language are overcome, the definition of the problem to everyone's satisfaction continues to be a problem. This is because the order and priorities of the knowledge very often do not mean the same things to the producer and the technologist.

For example, if we want to introduce a new product (say in Latin America or Asia), the producer will worry more about what new product he should put on the market and why rather than about how the product will be made. The technologist is usually more concerned about how to make it (1).

The producer (at industrial and artisanal levels) is not worried about how to make the new product because he instinctively assumes technology is available (very often for free). And why not, if the extensionist is right there to offer him the technological knowledge.

What mostly interests the producer is the forecast of economic, legal, cultural, psychological, environmental and, quite often, the political consequences of a projected technological change (8). Not only are technology and benefits derived therefrom important but other factors can equally influence the final decision.

One recurring example of this is the failure to introduce new fish handling methods on board, which can lead to increased benefits through better quality, a reduction in loss, etc. The technology is available but it is very difficult to put into practice. Usually the crew is reluctant to change their working habits even if they are paid more for doing the same job. Once

you are able to convince the crew that the change means also less work, you can proceed from there and try to explain to the owner of the boat why he should continue to employ the same number of people on board and pay them the same salary as before for doing less work.

Another example are the hundreds of fishing boats rusting in harbours because someone decided that it was the best technology to introduce. In this case, the legal, economic and political decisions were made to suite the technology already chosen. Of course, the whole system collapses when the situation demands a change in economic or policy rules. This kind of technological development rarely lasts and very often a bad memory of past experience will hinder the introduction of a really appropriate technology.

So the extensionist should not be too surprised when he offers his services to the industrialist and is told "You can really help me by convincing the government to lower its taxes"; or to the small fishmonger and is told "You can really help me by convincing the inspectors to apply less rigid fish controls". These are, of course, two extreme examples (although taken from real instances).

Lack of Confidence

As in other types of communication, technological communication should be played with a minimum of confidence in others.

Industrialists in developing countries usually take it for granted that research institutes and technologists in their own countries are useless. Even at artisanal level the foreign adviser often succeeds where the national adviser fails. The success or failure does not depend only on the differences in knowledge or experience but is mainly based on the fact that one was a foreigner and the other a national. It is also a literal translation of the biblical proverb that no one is a prophet in his own land.

On the other hand, technologists frequently classify their own industry as being "primitive", "backward" or "underdeveloped", which is definitely not a good base for starting communication.

Even when a degree of mutual respect is achieved, the technologists may tend to think that it took too much time for industry to adopt new techniques or, adversely, the industrialists may tend to think that too much time was spent on research that may turn out to be useless when there are so many unsolved day-to-day problems.

Of course, there is some justification on both sides. Industry cannot be constantly changing techniques. They must be reassured that each new technique is economically beneficial to industry and this assurance takes time, say five or ten years. Pure research rarely takes into account financial constraints.

On the other hand, technologists working at universities or in research and development institutes are seldom evaluated by their direct contributions to industry. More common, they receive recognition by the number and/or quality, measured in terms of "excellence", of publications they produce (the peer's judgement). In this situation, it is not surprising if they orientate their work towards goals that will facilitate their careers.

Finally, there is very often a deep conviction that it would be a waste of time to sit down all together and discuss common points of interest. Each side considers the other side hopeless.

My personal opinion is that technologists (extensionists or researchers) should take the first step. Producers at industrial or artisanal level are usually involved in a more or less difficult day-to-day struggle to carry out their activities whereas the technologists are not under the same pressures.

The Need for Dialogue Between Effective Spokesmen

Development, amongst other things, is a result of organization. But development in turn generates new demands on organization. As a result, development in any area of technology implies in practice a division of work.

In developed countries each division is a consequence of the need to make the production system in each specific area more efficient. In practice, there is (or should be) a connecting link between the different areas to ensure the smooth running of the whole.

Large companies in developed countries even have their branch of R&D which means they also take part in promoting a technology of their own. Middle sized companies frequently have a person in charge of technical and scientific matters. Small companies join an association which looks after their technological interests. Developed countries also have intermediate companies which merchandise in technological knowledge. There is also a large circulation of journals and magazines devoted to specific technical subjects and many occasions for industrialists and researchers to get together, e.g. fairs, technical congresses, etc. (7).

Developing countries do not have the same advantages, irrespective of the size of their industry or academic level of the universities and research institutes.

Whereas in developed countries it is not unusual for university professors and senior researchers to advise the industry (10)(2), in developing countries not only is this not done but quite often it is frowned upon.

Some Latin American countries, in particular, have regulations to control universities in their contacts with industry that in

practice prevent communication. Advising industry is not considered part of technology self development but as some kind of business with a high risk of becoming immoral (sometimes it is also illegal). The researchers and university professors are seen as medieval monks in secular and contemporary clothes who should remain "pure" from external contamination. This reminiscence of religious (medieval) universities, still alive in South American universities, will stay alive until it is discovered that it is less harmful and more beneficial for the country to allow researchers and professors to advise industry (and make some extra money).

In many developing countries the roles of technologist, researcher (in technology), extensionist and professor (in technological subjects) are combined. In other words, there are not several people doing several different jobs but only one person doing everything. Say you are appointed researcher. You are not only expected to produce new ideas for publication in international journals but you are also expected to train others on a regular basis and perform extension successfully (without infringing the regulations).

Institutes in developing countries should have a specific branch to handle communication with industry. It need not be four or five different branches but they should have an effective spokesman whose sole purpose is to perform this function.

Industrial associations should take a more active interest in technological aspects which have an effect on them. These associations could very well become the intermediary bodies between technological institutes and industry. They could promote knowledge and at the same time make the technologists aware of the problems facing industry.

## DISCUSSIONS AND CONCLUSIONS

This paper is a review of the most important problems hindering the internal transfer of technology in developing countries, mainly in the Latin American region. It suggests ways of initiating or improving communication between industry and institutes.

A detailed analysis of all the problems associated with communication is beyond the scope of this paper. Many important factors such as economic and financial constraints, influence of the research pattern chosen by the country, institutes' creativity and organization, cultural influences, marketing and distribution problems, role of external aid, etc., were only mentioned briefly. The intention was to show that the problem of technical communication in developing countries is more complex than usually thought.

It is also clear that a mechancial approach to communication, e.g. trying to convince people through carefully planned demonstrations, is not sufficient in developing countries. The necessity for extension services has been underlined and the attitude and philosophy of these services outlined.

Probably the best recommendations are to link the observations made in this paper with the concept of technology assessment as applied to the food industry (1)(8) and the economic analysis of proposed technological changes.

Of course, the magnitude of each problem will dictate the extent of the assessment necessary. Again, it is advisable that institutes wishing technological communication first organize a multi-disciplinary team to work and advise on a regular basis the whole question of assessing the internal transfer of technology.

Universities and research institutes dealing with fish technology in Latin America and other developing countries in Asia are in a position to actively participate through technical communication with the processors in the improvement and development of fish utilization in their own countries. In order to do so, they should review their attitudes and ideas regarding fish processors and analyse their respective relationships.

Leonardo da Vinci wrote "The most useful kind of science will be that whose fruits are more communicable, and contrarily, the less useful will be the less communicable" (6). It is not a bad idea to remember this from time to time.

## REFERENCES


1. ALBRECHT, J.J. 1982. Technology's role in product development. Food Technology, Vol. 36(9), pp. 73-76.

2. ANON. Fall 1982. Helping the seafood industry control quality. The University and the Sea. Texas A&M University. pp. 2-4.

3. FREIRE, P. 1979. Extensao ou comunicacao? Ed. Paz e Terra, 4a. Edicao (Brazil).

4. JAMUL, D. 1981. A microlevel view of obfuscation. Ceres No. 84(14). pp. 39-40.

5. JAMUL D. 1981. Plain talk. Clear communication for international development. Ed. by Volunteers in Technical Assistance (VITA), Md., U.S.A.

6. LEONARDO DA VINCI. 1947. Tratado de la pintura. Espasa Calpe Arg. 2da. Edic.

7. LUPIN, H.M. 1982. Problems regarding extension of fisheries technology in developing countries. Proceedings of the Seventh Annual Tropical and Subtropical Fisheries Technology of the Americas. TAMU-SG-82-110. April.

8. MAING, I.V. 1982. The role of technology assessment in food research and product development. Food Technology, Vol. 36(9). pp. 65-66.

'9. POLINAC, R.B. 1978. Sociocultural factors influencing success of intermediate food technology programs. Food Technology, Vol. 32(4). pp. 89-92.

10. REGENSTEIN, J.M. and O.R. NOYES. 1982. University Involvement in the Commercialization of new products. Food Technology, Vol. 36(9). pp. 86-88.

11. WIONCZEK, M.S. 1977. La transferencia de tecnologia contemplada como proceso social. Interciencia, Vol. 2(5). pp. 262-263.

# TEMPORAL CHANGES IN FISH COMMUNITY DIVERSITY NEAR A SEWAGE OCEAN OUTFALL, MŌKAPU, O'AHU, HAWAI'I

ANTHONY R. RUSSO

WATER RESOURCES RESEARCH CENTER
UNIVERSITY OF HAWAII, HONOLULU

## INTRODUCTION

Although the field survey demands much effort, it is the most direct method of appraising long term ecological response. Glover, Robinson, and Colebrook (1972) have noted that the present understanding of variability in the field and the present level of monitoring of the natural environment are inadequate for the detection and identification of the sources of variation. It is difficult to separate natural processes from all but the most obvious pollution incidents.

Studies of the effects of sewage effluent on receiving waters close to outfall sites have proliferated at an accelerated rate during the past 10 years because of interest by local, state, and national governments in environmental quality. Observations on California outfalls made by Eppley et al. (1972) and Turner (1965) showed some evidence of eutrophication and large changes in relative abundance of organisms associated with this condition. The damage suffered by marine life when discharge is voluminous and dispersion conditions are not favorable was investigated by Bellan (1970) in France; environments changed as a result of sludge deposited over long periods of time have been described by Pearce (1970). Chen and Orlob (1972), McIntyre and Johnston (1974) and Topping (1976) all comment on the effects of sewage effluent on receiving waters in the coastal areas of the U.S.

Sewage outfalls present special problems to the investigator both in field survey technique and in the interpretation of the data. The efficacy of using baseline studies, once thought to be essential for environmental impact work, is now being questioned (Gray 1976). With regard to temporal variation, it is often recommended that a baseline be established where no effluent is present. Lewis (1971) has repeatedly pointed out that baselines may be particularly difficult to establish because long-term fluctuations characteristic of natural marine communities may greatly exceed those which are apparent in pre- or post-event studies.

For the investigator interested in ecological responses to sewage stress biological indicators are important (Word, Myers, and Mearns 1977). Indicator organisms are primarily used to identify rather than to measure environmental changes. The cause of these changes may remain unknown. Tolerant indicator species can direct attention to pollution because of large increases in their relative abundance. The absence of sensitive species may indicate some stress in the environment, and changes in species diversity and population density in apparently suitable ecosystems may serve as guides to ecosystem stress from a wide variety of sources. However, the use of indicator species is hampered by

our lack of knowledge of their biology and their normal population fluctuations in the natural environment. It may also be unsuitable to use changes in species diversity since we know very little about natural variations in relative abundances of species due to changes in fecundity, growth dynamics, and abiotic and interactive environmental factors. It is the exception rather than the rule when changes in species composition or community diversity can be related directly to man-made pollution. Yet, in order to understand the consequence of pollution, we must continue to study changes in the environment by using the best tools at our disposal. The use of indicator organisms as tools is one of the best options we have at the present to establish the presence of pollutants and the extent of their effects.

There is always a temptation to subject relatively straightforward sets of data to cumbersome statistical analysis. Quantification is necessary and desirable only insofar as it can be linked to biological processes. In this study the similarity is measured between fish communities at the same location at different sampling times in order to assess any changes in the surrounding environment due to outfall performance. Quantitative calculations of similarity and diversity between communities plays an important role in statistical ecology. Useful ways of measuring differences in similarity indices is to compare those which discriminate between dominant and rare species (Morisita 1959) and those which just measure presence or absence where each species contributes equally (Sorensen 1954). The efficacy of using either method is ultimately left to the investigator and should be mitigated by the problems to be solved. Grassle and Smith (1976) have developed a unified approach to similarity by defining a family of measures in which the contribution of rare and dominant species to each measure is explicit.

In this study the fish communities at various sites near a sewage outfall were monitored over a period of years to assess the extent of changes, if any, in the surrounding environment. A study of the benthic and fish communities was conducted at Mōkapu, O'ahu, Hawai'i (Russo, Dollar, and Kay 1977) in December 1975 prior to construction of an ocean outfall.

The outfall which discharges 0.39 $m^3/s$ (9 mgd) of secondarily treated sewage was constructed by the City and County of Honolulu and began operation in December 1977. The total outfall length is 1 500 m, including the diffuser which has a length of 293 m and lies in 26 to 31 m of water. Investigations were repeated at the same sites in the summer of 1978 (Russo, Dollar, and Kay 1979) and again in August of 1979.

## DESCRIPTION OF STUDY SITES

The outfall site is located south of Mōkapu Point off the north shore of O'ahu, Hawai'i (Fig. 1). Station B coincides with the Mōkapu Outfall; three Stations C, D, and E are located 1 600, 4 000, and 5 600 m south of the outfall. Station A is located 1 600 m north of the

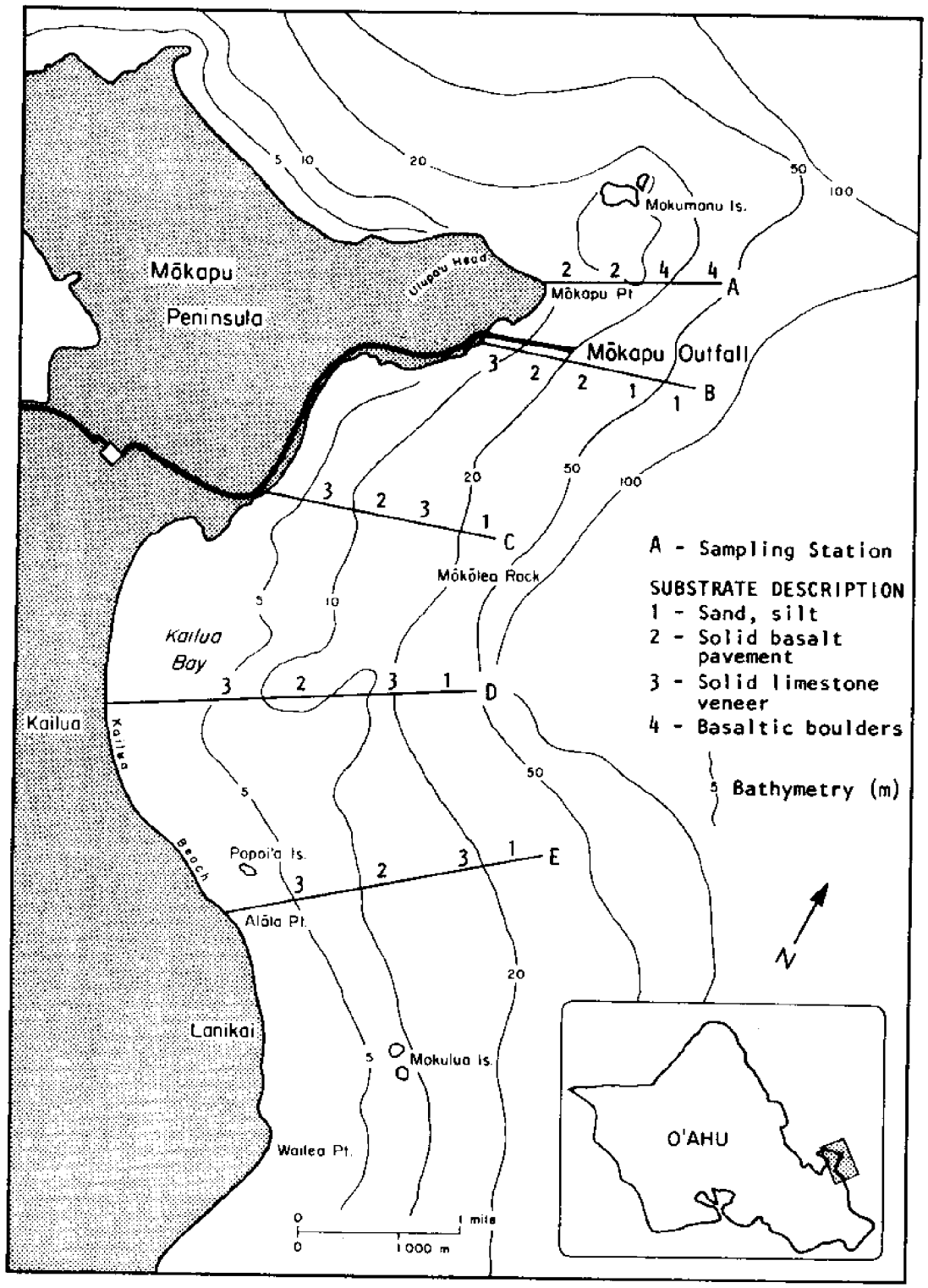



Figure 1. Sampling stations, Mōkapu, O'ahu, Hawai'i

outfall off Mōkapu Point, Station C is located north of Kailua Bay, Station D in Kailua Bay, and Station E off Alāla Point.

## METHODS AND MATERIALS

Using SCUBA, fish surveys were made at Stations A through E at depths of 6, 12, and 18 m (substations) in December 1975. June 1978, and August 1979. A 30-m transect line marked at every meter was laid on the bottom. A diver swimming along the line counted and identified fish species 3 m on either side of the transect line so that a 180 $m^2$ area was swept. At Station B a fish count was made at each depth on the outfall including one at 26 m and on the diffuser at 31 m. Fish species relative abundance (J) was calculated using the Shannon-Wiener index H' where J = H'/H' max. The number of species (richness) and their relative abundances were recorded for each fish count. To compare fish communities over time two similarity indices were used. The first, Sorenson's (1954) index (SI) measures the redundancy of fish species from one community to another in space or time. The equation for the index is SI = 2C/A + B where

C = no. of species in common to two transects at times $t_1$ and $t_2$
A = no. of species counted at time $t_1$
B = no. of species counted at time $t_2$.

Using the 50% rule (Mayr 1944) two aggregations of species occurring naturally in the same area at different times are to be considered distinct communities when at least 50% of the species in each aggregation are exclusive to the aggregation. Mueller-Dombois and Ellenberg (1974) suggest that an index of 50% (.5) represents a threshold value; that is, if the index exceeds .5 the similarity is great enough to indicate that the species are part of the same association or community. The second index of similarity (MI) used was derived by Morisita (1959). In this case

$$MI = \frac{2\sum_{i=1}^{K} n_i(t_1)\, n_i(t_2)}{[\lambda(t_1)\lambda(t_2)]N(t_1)N(t_2)}$$

where

K = species in common

$n_i(t_n)$ = no of fish in species i at time $t_n$

$N(t_n)$ = total number of fish counted at time $t_n$; and

$$\lambda(t_n) = \frac{\sum_{i=1}^{S} n_i(n_i-1)}{N(N-1)};$$

where

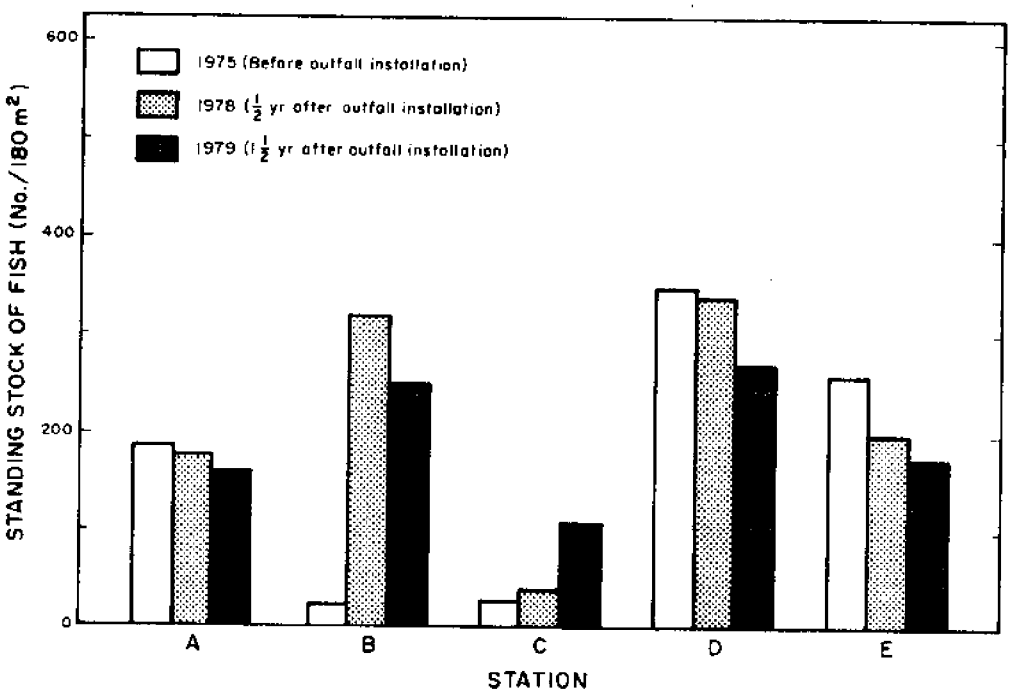
S = total no. of species at time $t_n$.

The 50% rule was also used with this index to determine the degree of similarity.

Photographs were taken at the outfall and the presence of macroinvertebrates was recorded. Using $\frac{1}{4}$-$m^2$ quadrats thrown randomly at each dive site, macroscopic algae were cropped and later, identified and weighed dry.

## RESULTS

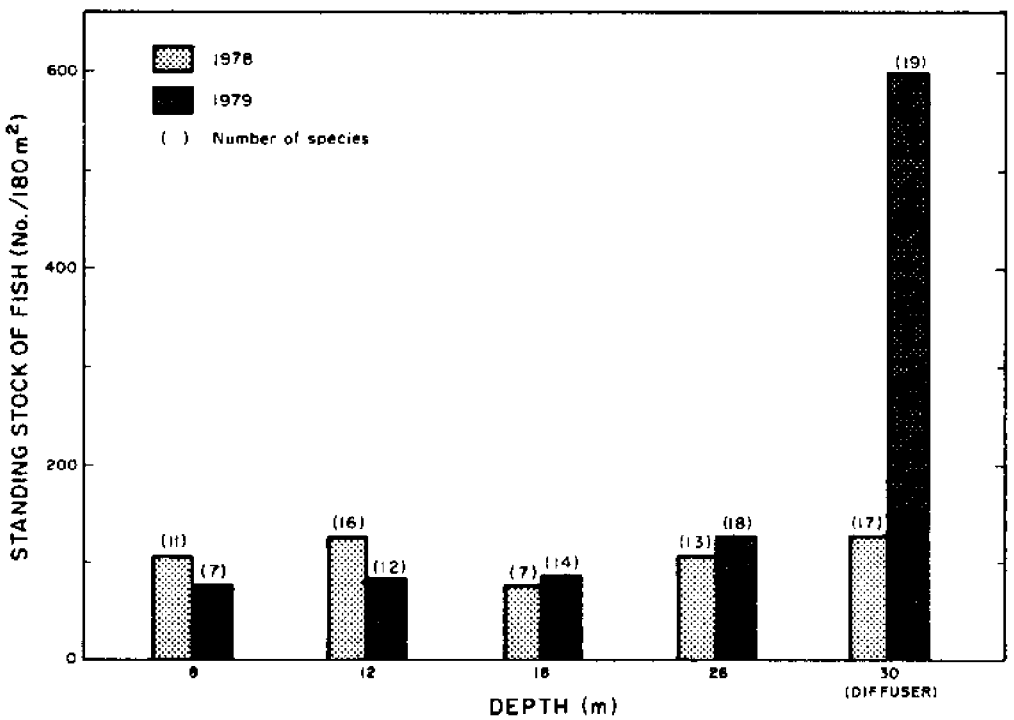
Figure 2 shows the comparative standing stocks of total fish counted at each station. Pre-outfall observations show that fish were scarce at both Stations B and C. Six months after outfall operation began (June 1978) there was a large increase in total fish at the outfall site (Sta. B), especially over the large rocks covering the outfall pipe. There were no significant differences in the total fish counted at the other stations after 6 mo of outfall operation. Eighteen months after the commencement of discharge (August 1979) at the outfall station large aggregations of fish were still present. The rudderfish Kyphosus cinerascens, the lemon butterfly fish Chaetodon miliaris, the goatfishes Mulloidichthys auriflamma and Parupeneus porphyreus, and the damselfish Abudefduf abominalis were seen along the outfall at depths from 18 to




NOTE: Outfall became operational in December 1977.

Figure 2. Comparative standing stocks of total fish at all stations, 1975, 1978, and 1979, Mōkapu, O'ahu, Hawai'i

26 m. The snapper Lutjanus kasmira was also seen at these depths. There were noticeable increases in fish abundance at Station C at this time. At the other stations (A, D, and E) there was a small percent decrease in total fish abundance (Fig. 2) from 1975 through 1979. At all substations except those at Station B similarity indices were high enough to indicate little change in community structure from 1975 to 1978 (Table 1). Where fish numbers and species richness were low in pre-outfall samplings they increased significantly in 1978 at the outfall (Sta. B). Fish of the families Acanthuridae, Pomacentridae, Mullidae, and Chaetodontidae were abundant. Results show little change in numbers of fish species and their relative abundance for all substations except for those at Station B after 6 mo of outfall operation. Between 1978 and 1979 there were no significant differences in fish community structure except at Station C (Table 1) and over the diffuser (31 m) at Station B. The relative abundances of fish counted in 1978 and 1979 over the outfall including the diffuser are shown in Figure 3. The abundance of fish over the outfall diffuser was significantly higher in 1979 than in 1978 due to very large aggregations (>500) of the snapper Lutjanus kasmira. This species was seen in 1978 but in much smaller numbers (<25). Changes in dominant feeding guilds over the outfall between 1978




NOTE: 90% of fish counted at 30 m in 1979 were of one species, Lutjanus kasmira.

Figure 3. Relative Abundance of Fish at Station B on the Outfall, Mōkapu, Hawai'i

Table 1. Comparative Data for Fish Populations from 1975 to 1979, Mōkapu Outfall, Hawai'i

| SUB-STATION | DEPTH | SIMILARITY INDEX | | | | % CHANGE IN H' | | % CHANGE IN TOTAL FISH† | | % CHANGE IN NO. OF SPECIES | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | S* | M | S* | M | | | | | | |
| | (m) | 1975-78 | | 1978-79 | | 1975-78 | 1978-79 | 1975-78 | 1978-79 | 1975-78 | 1978-79 |
| A-20 | 6 | .61 | .42 | .70 | .75 | -12 | -3 | -8 | -1 | -20 | -18 |
| A-40 | 12 | .60 | .57 | .45 | .77 | -19 | +14 | -3 | +7 | -17 | +10 |
| A-60 | 18 | .67 | .59 | .73 | .85 | -2 | +1 | -22 | +.1 | +4 | +1 |
| Off Outfall | | | | | | | | | | | |
| B-20 | 6 | .25 | .10 | .40 | .78 | +123 | +3 | +82 | -63 | +77 | -37 |
| B-40 | 12 | No fish counted | | .40 | .75 | --- | +30 | +100 | +37 | +100 | +200 |
| B-60 | 18 | No fish counted | | 0 | --- | --- | One species seen in 1978 | +100 | +350 | +100 | +500 |
| On Outfall | | | | | | | | | | | |
| B-20 | 6 | --- | --- | .67 | .77 | --- | -7 | --- | -42 | --- | -36 |
| B-40 | 12 | --- | --- | .50 | .73 | --- | -7 | --- | -35 | --- | -25 |
| B-60 | 18 | --- | --- | .32 | .58 | --- | +27 | --- | +8 | --- | +100 |
| B-80 | 26 | --- | --- | .46 | .59 | --- | +8 | --- | +12 | --- | +13 |
| On Diffuser | 31 | --- | --- | .45 | .37 | --- | -47 | --- | +600 | --- | +10 |
| C-20 | 6 | .8 | .71 | .41 | .71 | +30 | +300 | +5 | +300 | +100 | +250 |
| C-40 | 12 | .55 | .68 | .40 | .42 | +25 | +40 | +11 | +12 | +16 | +40 |
| C-60 | 18 | .55 | .70 | .31 | .23 | +11 | +28 | -28 | +73 | +8 | +30 |
| C-20 | 6 | .55 | .68 | .50 | .69 | +0.5 | -12 | -14 | -20 | +20 | -25 |
| D-40 | 12 | .61 | .71 | .53 | .73 | -12 | -8 | -28 | +8 | -5 | -10 |
| D-60 | 18 | .51 | .62 | .63 | .78 | +1 | -14 | +25 | -19 | +20 | -28 |
| E-20 | 6 | .45 | .60 | .50 | .65 | -7 | +2 | -7 | -17 | -16 | +15 |
| E-40 | 12 | .57 | .70 | .57 | .71 | -5 | -12 | -16 | -2 | -10 | -15 |
| E-60 | 18 | .58 | .65 | .50 | .70 | -11 | +10 | -20 | -9 | -17 | -13 |

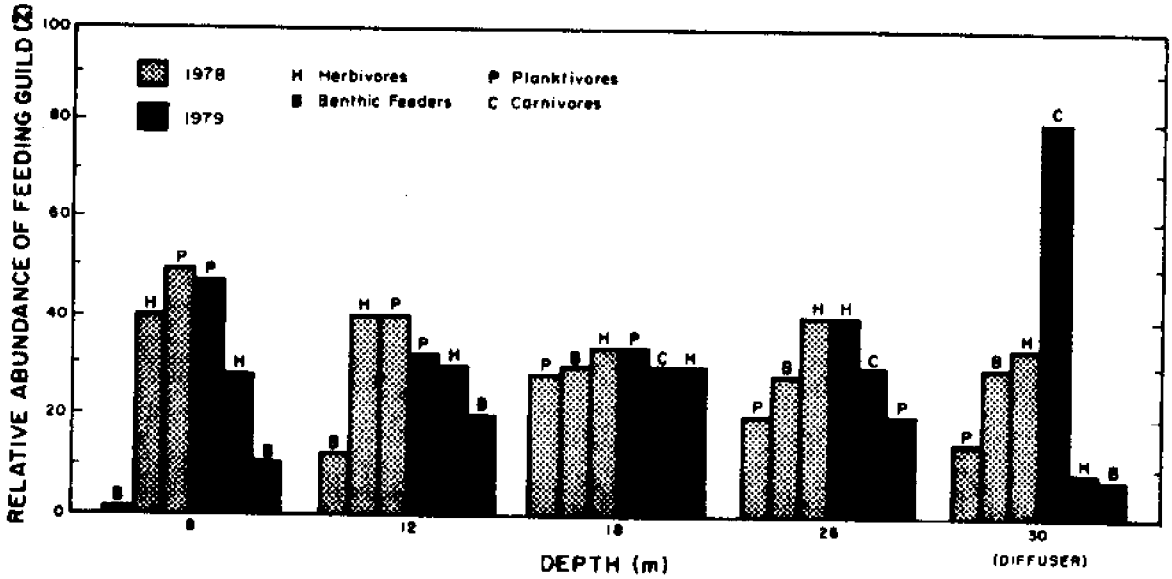



Figure 4. Dominant feeding guilds for fish communities on Mokapu, Hawai'i outfall site (Station B)

and 1979 samplings are shown in Figure 4. There were no significant differences in trophic feeding levels at 8 and 12 m. However, at greater depths significant changes in fedding guilds were observed. At 18 m carnivores became dominant in 1979 replacing herbivorous and benthic feeding fishes which were dominant in 1978. In 1979 at 26 m large numbers of carnivorous fish (the snapper Lutjanus kasmira) appear replacing benthic feeders, mainly goatfishes and wrasses, which were dominant in 1978. In 1979 at the diffuser (31 m) 80% of the fish counted were the snapper Lutjanus kasmira. In 1978 small numbers (<20) of this species were seen around the diffuser.

There is no significant difference in feeding guild dominance at most other substations between 1978 and 1979 (Table 2). Lutjanus kasmira, however, does show up at Substation D-60 (18 m) in 1978 and 1979. Planktonivorous fishes are dominant only over the outfall and at Station D (Table 2).

In 1975, 1978, and 1979 results of algae collections showed insignificant amounts of algae at most substations (<20 g dry/$m^2$). In 1979 moderate amounts of algae (90-180 g dry/$m^2$) of the general Dictyopteris and Halimeda were seen at depths of 12 and 18 m at Station C. There was no significant difference between the biomass of algae seen at these substations for all three sampling times (Russo, Dollar, and Kay 1979, Table 2). On the rocks over the outfall pipe at depths of 26 and 31 m significant amounts (>250 g/$m^2$ dry) of the filamentous blue-green alga Lyngbya were collected. Herbivorous fishes were observed feeding on this alga which grew approximately 5 m on either side of the outfall in depths from 18 to 26 m. This alga was not recorded in 1978. In 1979

Table 2. Percent Relative Abundance of Dominant Feeding Guilds, Mōkapu Point, O'ahu, Hawai'i, 1975-1978

| SUB-STATION | DEPTH | PRE-OUTFALL | | | 6 MO POST OPERATION | | | | 18 MO POST OPERATION | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Dominant Feeding Guilds* | | | | | | | | | | |
| | | H | B | P | H | B | P | C | H | B | P | C |
| | | (% Relative Abundance) | | | | | | | | | | |
| A-20 | 6 | 42 | 25 | -- | 50 | 30 | -- | -- | 43 | 33 | -- | -- |
| A-40 | 12 | 22 | 25 | -- | 15 | 70 | -- | -- | 18 | 22 | 18 | -- |
| A-60 | 18 | 17 | 33 | 33 | 30 | 33 | 13 | -- | 33 | 30 | 33 | -- |
| A-80 | 26 | 20 | 55 | -- | 25 | 44 | -- | -- | 20 | 50 | -- | -- |
| Off Outfall | | | | | | | | | | | | |
| B-20 | 6 | 50 | 50 | -- | 25 | 65 | -- | -- | 20 | 66 | -- | -- |
| B-40 | 12 | No fish seen | | | -- | 100 | -- | -- | 10 | 72 | -- | -- |
| B-60 | 18 | No fish seen | | | 37 | 31 | 21 | -- | 40 | -- | 20 | 30 |
| On Outfall | | | | | | | | | | | | |
| B-20 | 6 | -- | -- | -- | 40 | -- | 55 | -- | 12 | 30 | 50 | -- |
| B-40 | 12 | -- | -- | -- | 40 | 10 | 40 | -- | 35 | 20 | 30 | -- |
| B-60 | 18 | -- | -- | -- | 35 | 30 | 28 | -- | 31 | -- | 34 | 32 |
| B-80 | 26 | -- | -- | -- | 37 | 31 | 21 | -- | 40 | 20 | -- | 30 |
| Diffuser | 31 | -- | -- | -- | 33 | 30 | 15 | -- | 10 | 10 | -- | 80 |
| C-20 | 6 | -- | 75 | -- | 50 | 50 | -- | -- | 50 | 50 | -- | -- |
| C-40 | 12 | -- | 80 | -- | 60 | 35 | -- | -- | 25 | 65 | -- | -- |
| C-60 | 18 | -- | 80 | -- | 50 | 50 | -- | -- | 20 | 55 | -- | -- |
| D-20 | 6 | 30 | 50 | 15 | 60 | 35 | -- | -- | 45 | 50 | -- | -- |
| D-40 | 12 | 44 | 33 | -- | 60 | 30 | -- | -- | 50 | 40 | -- | -- |
| D-60 | 18 | 25 | 10 | 50 | 33 | -- | 28 | 33 | 38 | -- | 50 | 10 |
| E-20 | 6 | 43 | 40 | -- | 66 | 20 | -- | -- | 35 | 33 | -- | -- |
| E-40 | 12 | 66 | 23 | -- | 58 | 36 | -- | -- | 38 | 28 | -- | -- |
| E-60 | 18 | 50 | 36 | -- | 50 | 25 | -- | -- | 40 | 55 | -- | -- |

*From Hobson (1974).
H = Herbivore
B = Benthic Feeder
P = Plankton Feeder
C = Carnivore (free swimming prey).

large aggregations (5-8 per $m^2$) of the echinoid Echinothrix spp. were counted along the outfall and under the rocks covering the outfall pipe. This represents a significant increase in sea urchin population since 1978. Small coral heads (5-10 cm in diam) of the species Pocillopora meandrina were seen on the rocks over the outfall pipe in both 1978 and 1979. Near the diffuser clumps as large as 20 to 30 cm of the bryozoan

Triphyllozoon hirsutum were seen in both 1978 and 1979 samplings.

## DISCUSSION

There were essentially no changes in the fish community species structure at Stations A, D, and E 6 and 18 mo after commencement of outfall operation (Table 1), and the feeding trophic levels remained essentially the same (Table 2). Fish counts at these stations were down from 1975 to 1979 but not significantly (Fig. 2). Communities would only be expected to change their species composition and relative abundance under extreme stress, e.g., depletion of feed supply, destruction of habitat space, or changes in the physicochemical environment. At this time the discharge from the Mōkapu Outfall does not appear to affect these areas. At Station C, however, there is a moderate increase of fish species and abundance in 1979 (Fig. 2). Similarity coefficients are low indicating a change in community structure from 1978 to 1979 (Table 1). This increase in species diversity and abundance may be due to increased migration of fish from the outfall site 1 500 m away. The southerly movement of diluted effluent with seasonal current drift (Bathen 1978) may enhance the food supply or conditions for growth of algae, the source of food for grazing fish. Results show that benthic feeders dominate at Station C but also that the relative abundance of herbivorous fishes increased after outfall operation commenced (Table 2). In general the biomass of algae did not change significantly during the 1-1/2 yr of outfall operation at most stations (Russo, Dollar, and Kay 1979). Results of algal harvests in 1979 showed little or no change from 1978. Most of the biomass recorded (80 g dry) appeared at substations C-40 and C-60. In 1978 at these 2 substations Dictyopteris and Halimeda were the only two genera seen but in 1979 patches of Sargassum, Asparagopsis, Lyngbya, and Wranglia were also recorded. However, there were significant amounts of Lyngbya seen at depths between 18 to 31 m on the outfall in 1979. This blue-green, nitrogen fixing alga was not seen in 1978 6 mo after outfall operation. According to Bohlool (1978) since nitrogen fixation is an energy requiring process, one would expect high fixation rates in areas receiving effluents rich in utilizable organic carbon and, if the discharge is low in nitrogen content, it would be reasonable to expect nitrogen fixing organisms to have a selective advantage.

The nitrogen levels at Mōkapu were sampled 2 to 3 mo after commencement of discharge (Laws 1979). The results of this sampling showed that nitrogen levels ($NO_2$ + $NO_3$) at the outfall site were three times higher after operation than before. Although concentrations of nitrogen compounds are high in the immediate vicinity of discharge, microorganisms—especially phytoplankton—rapidly assimilate these compounds (Ryther and Dunstan 1971). Sewage effluent is phosphate rich; in the nitrogen deficient receiving waters blue-green algae can utilize $PO_4$ while fixing their own nitrogen compounds.

At the outfall site (Sta. B) similarity indices show that 6 mo after the commencement of outfall operation there was a significant in-

crease in the fish community diversity due to large increases in species richness and abundance (Fig. 2). The large rocks covering the outfall attracted large numbers of fish due probably to the increase in attachment sites and habitat space for marine organisms. The rudderfish (Kyphosus cinerascens) was abundant in 1978. This fish, a strict herbivore, was seen grazing on attached filamentous algae at depths of 12 and 18 m on the outfall. Acanthurids, which are herbivores in general, were also abundant along the outfall. Large numbers of goatfish (Mulloidichthys auriflamma) were observed feeding in the sediment along the outfall pipe at depths of 6, 12, and 18 m. Goatfish normally feed on benthic infauna and at times on organic detritus in the sediment (Hobson 1974). Whatever their food source the outfall attracted them in large numbers. These observations are consistent with other studies done on outfalls (Turner 1965) in which large increases of fish were associated with outfall construction. One year later (1979) results show that, except on the diffuser itself, there was no change in fish community structure over the outfall (Table 1) and the feeding trophic levels of the fish community remained the same (Table 2).

Generally plankton feeding fish were dominant over the outfall. Many fish are adapted to foraging for plankton and organic particles in a water column above a reef substrate (Davis and Birdsong 1973). Large aggregations of the planktonivorous damselfish Abudefduf abdominalis were seen in the water column above the outfall. New substrate afforded by the construction of outfalls are ideal for water column foragers since there may be a constant influx of organic particulate matter from below.

Table 2 shows that there was an increase in carnivorous fishes (those feeding on free swimming prey) at substations B-60 and B-80 due mainly to the appearance of the snapper Lutjanus kasmira. The number of herbivorous fishes decreased in deference to this species. Results of dives on the diffuser show a large increase in total fish and a slight increase in species richness from 1978 to 1979 (Fig. 3). Eighty percent of the fish counted were the carnivore Lutjanus kasmira (Fig. 4). Large numbers of surgeonfish (Acanthuridae), goatfish (Mullidae) and wrasses (Labridae) were also seen. The snapper taape (Lutjanus kasmira) was introduced to Hawai'i in the mid '50s essentially as a potential food fish. It has been reported in large numbers off the coast of O'ahu by many divers. Being a predator of small free swimming prey (Hobson 1974) it may put great pressure on the juvenile fish stocks of other species seen at the diffuser and could cause a sharp decline in diversity and abundance of fish there. The total landings of taape from all the islands have increased from 1000 pounds in 1967 to 100,000 pounds in 1981 netting about $70,000 (Tabata 1981). This fish seems to disperse rapidly and to quickly exploit new habitat space. There have been dramatic increases in sighting by divers and fishermen report significantly higher catches in recent years than normal. Even though taape is fished it is considered of secondary importance as a commercially valuable fish. It is only taken when other more commercially valuable fish are absent. Increases in taape have been reported by local fishermen in areas where

they also complain of declines in preferred species such as goatfish (Mulloidichthys and Parupeneus), big eye (Priacanthus), and squirrelfish (Myripristus). There is no scientific evidence to indicate that taape is outcompeting or overlapping in niche with these other species. However, stomach contents do indicate that the taape may be a general carnivore taking, along with crustaceans, juveniles of the abovementioned species. There is no indication that taape is eaten by its cohabitors (Tabata 1981). Whether or not there is a reciprocal density dependent relationship between taape and its cohabitors is not clear but a definite increase in abundance of this fish over the last 20 years and its rapid dispersal to all islands is well documented. The low demand for taape by consumers and fishermen simply lies in the fact that it is a colorful yellow fish with pale flesh. Local fish consumers traditionally prefer "red" fish. Most consumers who try taape consider it as tasty as the other commercially valuable reef and shore fish. The University of Hawaii Sea Grant Program has begun a campaign to "re-educate" fishermen and consumers about the palatability and market value of taape. Progress is slow, traditions die hard as most fishermen still consider the taape as a "junk" fish (Tabata 1981). If these cultural biases can be removed and the demand for taape increased, along with its price per pound, the culling of this species will not only be of commercial value but ecologically efficacious since it will insure the stability and diversity of the fish community. Since sewer outfalls attract large aggregations of taape and its cohabitors, these areas could be used as fishing grounds (Russo 1979). The taape is easily caught with gill nets and purse seines and Hawaiian sewer outfalls are at present at depths between 30 and 60 meters (100-200 ft) making it easy for fishermen to work. Preliminary observations on a new sewer outfall at Barbers Point, O'ahu in 60 meters of water show large aggregations of taape at the site along the pipe (Russo et al. 1981).

Although there were significant increases in filamentous alga (Lyngbya) along the diffuser and an increase in detritus feeding echinoids, there seems to be no buildup of sewage sediment. This is corroborated by the fact that colonies of the filter feeding bryozoan Triphyllozoon hirsutum (lace coral) seen in 1978 are still present at the diffuser in 1979. The Mōkapu Outfall diffuser seems to be an ideal habitat for this bryozoan. The organic load of the surrounding water is high enough to provide abundant material for filter feeding while on the other hand the rapid dispersion of the effluent by water movement keeps the organic load low enough to prevent destruction of the bryozoans.

## SUMMARY

An analysis of fish community similarity in the area of the Mōkapu ocean sewage outfall show no widespread adverse changes in the surrounding environment from outfall operation over a 1-1/2-yr period. In general fish communities maintained their structure and showed little change in diversity over the sampling period. The clarity of the receiving waters and adjacent areas is good and the presence of filter

feeding bryozoans may indicate sufficient dispersal of sediment by vigorous water movements at the outfall site.

The abundance and diversity of fish generally increased near the outfall as a direct result of new substratum afforded by outfall construction. At a station 1 600 meters south of the outfall increases in fish species richness and abundance were observed. There was a large increase in relative abundance of one species of carnivorous snapper (Lutjanus kasmira) near the outfall. This species was introduced for its commercial value in 1955 but has not become a popular food fish due to cultural biases. Its rapid increase in abundance and its rapid dispersal to all islands makes it a good candidate for fishing especially at newly constructed outfall sites where they aggregate being attracted by new habitat space and prey. Culling this population will be of commercial value but more importantly could be the only population control mechanism due to the absence of any known predators.

There were no significant increases in the biomass of macrophytes except near the outfall where the blue-green alga Lyngbya is dominant. This may indicate an enrichment of the receiving waters with phosphate since nitrogen fixers respond well in phosphate rich waters with high C:N ratios (Bohlool 1978).

## REFERENCES


Bathen, K.H. 1978. Circulation atlas for Oahu, Hawaii. UNIHI-SEAGRANT-MR-78-05, Sea Grant College Program, University of Hawaii.

Bellan, G. 1970. Pollution by sewage in Marseilles, France, Mar. Poll. Bull. 1:59-60.

Bohlool, B.B. 1978. Nitrogen fixation in polluted intertidal sediments of Waimea Inlet, Nelson. N.Z. J. Mar. and Freshwtr. Res. 12(3):271-75.

Chen, C.W., and Orlob, G.T. 1972. The accumulation and significant of sludge near San Diego outfall. J. Water Poll. Control Fed. 44:1362-71.

Davis, W.P., and Birdsong, R.S. 1973. Coral reef fishes which forage in the water column. Helgolander Wiss. Muresunters 24:292-306.

Eppley, R.W.; Carlucci, A.F.; Holm-Hansen, O.; Kiefer, D.; McCarthy, J.J.; and Williams, P.M. 1972. Evidence for eutrophication in the sea near southern California coastal sewage outfalls. Rep. 16, Calif. Co-Op. Oceanic Fish Invest., p. 74.

Glover, R.S.; Robinson, G.A.; and Colebrook, J.M. 1972. Analysis of long term variation in composition and abundance of plankton in the North Atlantic. In Marine Pollution and Sea Life, ed. R. Johnston, p. 404. London: Academic Press.

Grassle, F.J., and Smith, W. 1976. A similarity measure sensitive to the contribution of rare species and its use in investigation of

variation in benthic communities, Oecologia 25:13-22.

Gray, J.S. 1976. Are baseline surveys worthwhile? New Scientist 70:219-21.

Hobson, E.S. 1974. Feeding relationships of teleostean fishes on coral reefs in Kona, Hawaii. Fishery Bull. 72:915.

Laws, E. 1979. Response of phytoplankton communities at Sand Island and Mōkapu Pt. to sewage discharges. Report prepared for City and County of Honolulu, Hawaii.

Lewis, J.R. 1971. Effect of crude oil and oil spill dispersant on reef corals. Mar. Poll. Bull. 2:59-62.

Mayr, E. 1944. Wallace's line in light of recent zoogeographic studies. Quart. Rev. Biol. 19:1-14.

McIntyre, A.D., and Johnston, R. 1974. Effects of nutrient enrichment from sewage in the sea. In Int. Symp. Discharge of Sewage from Sewer Outfalls. London, England, 19 August, No. 14.

Morisita, M. 1959. Measuring of interspecific association and similarity between communities. Mem. Fac. Sci., Kyushu Univ. Ser. E (Biol.) 3:65-80.

Mueller-Dombois, D., and Ellenberg, H. 1974. Vegetation ecology. New York: John Wiley.

Pearce, J.B. 1970. Long term effects associated with dumping of sewage sludge. In FAO Tech. Conf. on Marine Pollution and its Effects on Living Resources and Fishing, F.I.R.: MP/70/E-99, Rome 9-18.

Russo, A.R.; Dollar, S.; and Kay, E.A. 1977. An inventory of benthic organisms and plankton at Mokapu, Oahu. Tech. Rep. No. 101, Water Resources Research Center, University of Hawaii, Honolulu.

Russo, A.R.; Dollar, S.; and Kay, E.A. 1979. Ecological observations off the Mokapu, Oahu ocean outfall: A postinstallation study. Tech. Rep. No. 122, Water Resources Research Center, University of Hawaii, Honolulu.

Russo, A.R.; Heilfrich, P.; Dollar, S. 1981. Biomonitoring of Deep Ocean Outfalls, Oahu, Hawaii: Reconnaissance of Benthic and Fish Communities, Phase I. Preliminary report to Dept. of Public Works, City and County of Honolulu.

Ryther, J.H., and Dunstan, W.M. 1971. Nitrogen, phosphorous, and eutrophication in the coastal marine environment. (Science 17: 1008-13).

Sorensen, T. 1954. Adaptation of small plants to deficient nutrition and a short growing season. Botan. Tidsskr. 51:339-61.

Stein, J.E., and Denison, J.G. 1963. Limitations of indicator organisms. In Pollution and Marine Ecology, ed. T.A. Olsen and F. Burgess. New York: John Wiley and Sons.

Tabata, R.S. 1981 (July). Taape: What Needs to Be Done. Paper no. 46, Sea Grant Program, University of Hawaii.

Topping, G. 1976. Sewage and the sea. In Marine Pollution and Sea Life, ed. R. Johnston, Academic Press, London, p. 303.

Turner, C.H. 1965. The marine environment in the vicinity of the Orange County Sanitation District's (California) ocean outfall. California Fish and Game 52:28.

Word, J.Q.; Myers, B.L.; and Mearns, A.J. 1977. Animals that are indicators of marine pollution. In Annual Report, 1977, Coastal Water Res. Proj., El Segundo, California, pp. 199-206.

# PRELIMINARY RESULTS OF A STUDY OF SURVIVAL AND GROWTH OF CUT vs. HOOKED COMMERCIAL SPONGES IN THE FLORIDA KEYS

John M. Stevely and Don Sweat
Florida Sea Grant Marine Advisory Program
117 Newins/Ziegler Hall
University of Florida
Gainesville, Fl. 32611

## INTRODUCTION

Up until the 1940's, the sponge fishery was one of the most valuable fisheries in Florida. However, a combination of the sponge blights of 1939 and 1946 and the introduction of synthetic sponges resulted in reduction of the fishery to a small fraction of its former importance (1). In recent years increasing scarcity of sponges in the sponge beds off of Tarpon Springs has threatened the continued existence of the remaining Tarpon Springs sponge fleet. Although an exploratory sponge fishing survey of state territorial waters off of Monroe County (Florida Keys) Florida had indicated that commercial quantities of sponges were present (2), a Florida State Statute (F.S. 370.17 (2)) prohibits use of "deep sea diving apparatus to harvest sponges in this area". This Statute states ... "No person may use diving suits, helmets, or other apparatus used by deep sea divers, in taking commercial sponges from any of the waters within the territorial limits of this state". It was enacted to prevent damage to young sponges caused by heavily weighted divers stepping on them. The purpose of the law was not to prohibits reasonable methods of harvesting sponges, but to protect young sponges. While early sponge diving gear was of the deep sea variety, scuba or hookah gear did not become a viable method of extended underwater diving until many years after enactment of the law. Today, light hookah gear with rubber-soled, canvas athletic shoes has replaced the heavy helmet and lead-boot clad diver.

Consequently, The Florida Saltwater Fisheries Study and Advisory Committee recomended in 1982 that sponge diving be allowed in the state territorial waters off Monroe County. However, objections were still raised concerning the possiblity of overfishing due to increased fishing pressure following legalization of sponge diving. Therefore the present project

was undertaken for the purpose of establishing whether a change in harvesting technique -- cutting rather than tearing the sponge loose -- could insure sponge regeneration and thus reduce the possiblility of overfishing occuring. Sponges have remarkable regenerative ability, and if even a small quantity of sponge material is left attached to the substrate, the sponge may survive and grow back to a commercially valuable size.

## METHODS AND MATERIALS

During June 1983, a commercial sponge fisherman assisted us with locating a sponge bed off of Vaca Key (Marathon) in the Florida Keys. The site was marked with a bouy and Loran C coordinates recorded to insure relocation of the study site. The bottom of the site was marked off by laying out a grid of polyurethane line held in place by rebar stakes pounded into the substrate. Sponges were located by swimming along the line. Upon locating a sponge its position was marked by attaching a piece of brightly colored tape to the line. The position was also recorded on a plastic chart of the grid pattern. Large calibers were used to take two measurements of the sponge's diameter and the one measurement of the sponge's height.

Sponges were either cut loose using a large sharp knife or torn loose with a sponge hook on the end of a 3 foot handle. The sponge hook was of the type used by commercial sponge divers. After the sponge was removed, the type of method used was recorded and measurements taken of the remaining sponge base (if there was any left). The exact location of each sponge was marked by driving a rebar stake into the substrate. A numbered plastic bird band was wrapped around these stakes for identification. Also, in those cases where sponge base was left behind, the sponge base was marked by "sewing" a piece of monofiliment line through it and then attaching a plastic bird band. Several sponges were left intact but tagged with monofiliment as a control. To date, we have returned to the site twice (July, 1982 and Nov. 1983). So far data has only been collected on survival, the sponges had not yet grown enough to accurately measure regrowth.

## RESULTS & DISCUSSION

A total of 69 sponges were either cut (35) or hooked (34). Return trips to the site have been sucessful in relocating the sponges. On the first return trip (Aug., 1983) 60 sponges were relocated (87%). Only 40 sponges (58%) were relocated in Nov. due to bad weather and poor water clarity.

The data collected on survival indicate that there is a significant difference in survival of cut sponges compared to hooked sponges (Table 1). Data from both return trips indicate that 85%-90% of the cut sponges survived. Data on hooked sponges

is somewhat less consistent, survival being 60% in the June trip and then dropping to 32% in the November trip. A review of the field notes indicated that several hooked sponges recorded as alive in the June sample were extremely small, thin patches of sponge material. These sponges were not alive at the November sampling and consequently the (long term) survival of hooked sponges was considerably reduced.

The data on hooked sponges is in agreement with sponge hookers who claim that when hooking a sponge, a piece of sponge is often left behind which has the potential to grow back. However, the data collected in this study shows that this is not the case 100% of the time. If the sponge is torn "cleanly" from the substrate, the thin film of sponge material left behind will not survive.

Table 1. Survival (%) and sample size (N) of relocated sponges in August and November 1982 samples.

| | Aug. 1982 | Nov. 1982 |
|---|---|---|
| Cut | 90% (N=30) | 86% (N=21) |
| Hooked | 60% (N=30) | 32% (N=19) |

## REFERENCES


1. STEVELY, J.M., J.C. THOMPSON and R.E. WARNER. 1978. The biology and utilization of Florida's commercial sponges. Florida Sea Grant Technical Report No. 8. 45 pp.

2. SWEAT, D. and J.M. STEVELY. 1981. A report on the availability of commercial sponges within the gulf waters of Monroe County, Florida and the impact of commercial diving operations upon this resource. Gulf and South Atlantic Fishery Development Foundation, inc. Tampa, Fl. 26 pp.

# PRELIMINARY OBSERVATIONS FROM ON-BOARD HANDLING DEEP-SEA CRABS (GERYON SP.) HARVESTED FROM THE GULF OF MEXICO

Jeffrey J. Bellairs & W.Steven Otwell
Department of Food Science & Human Nutrition
University of Florida
Gainesville, Florida 32611

## INTRODUCTION

The possibility of initiating a new crab fishery in the Gulf of Mexico is generating considerable interest in the Gulf coast seafood industry. Fisherman searching for alternative fisheries and processors aware of the recent decline in king crab production are both interested in the development of this fishery. The potential species is a deep-sea variety which inhabits the Gulf at depths of 200 to 350 fathoms. It is a member of the genus Geryon and a species that is yet to be identified. Collaborative work with crustacean expertise at the Smithsonian Institute in Washington, D.C. has been arranged to provide a more current identification. Previous exploratory fishing in the Gulf region by the National Marine Fisheries Service during 1956 thru 1970 recorded two species, Geryon quinquedens and Geryon affinis (8). Both species are found along the eastern seaboard from Maine to Florida (4). These two species, primarily G. quinquedens, are termed red crabs and are currently harvested in New England.

Development of the New England red crab fishery, in the early 1970's, encountered high crab mortality rates during transport to dock and the formation of a black discoloration on the shell and flesh of butchered crabs. Holmsen (5) studied survival rates of red crabs directly iced and also packed in moist burlap bags and iced. High mortality rates were encountered in both storage methods and Holmsen and McAllister (6) recommended refrigerated sea water (RSW) storage. Meade and Gray (7) investigated the possibility of butchering red crabs on-board as an alternative to costly, non-dependable RSW storage. They found that the butchered crab halves developed a black discoloration that made the crab aesthetically unacceptable. Meade and Gray assumed the discoloration to be due to the action of a polyphenol oxidase enzyme commonly associated with "black spot" in shrimp. They found that the darkening could be inhibited for 19 days by dipping the butchered halves into a 1.00% sodium bisulfite solution for 10 seconds. Additional work has shown that multiple forms of a polyphenol oxidase enzyme do exist in G. quinquedens (3). Modern red crab boats are equipped for on-board butchering, cooking and

freezing, thereby alleviating the problems of live storage and discoloration.

Preliminary observations of the Gulf of Mexico deep-sea crab indicated that the problems of high mortality rates during transport and black discoloration were also common to this species. Therefore, the ongoing objectives of this study are to find an economical method of storage during transport from the fishing grounds, develop a procedure to retard the discoloration and to develop recommendations regarding storage life, butchering and cooking.

## MATERIALS AND METHODS

Gulf of Mexico deep-sea crabs were caught using various bottom traps, baited and strung in bottom longline fashion. Fishing was conducted during July and early September in the Gulf of Mexico, approximately 120 miles west of Tampa, in 200 to 300 fathoms. The gear and description of the harvest, including crab size and weights are provided by Sweat and Otwell (9). Experimentation, using typical crabs from this harvest, was conducted on-board and in the Department of Food Science and Human Nutrition, at the University of Florida, Gainesville.

### Survival Studies

Survival studies were carried out in the boat hold (50°F). Three separate samples, each containing 25 male and 25 female crabs, less than one hour out of the water, were either:

(i) directly held on top of ice;

(ii) separately packed into burlap bags moistened with sea water and iced;

(iii) or separately packed into plastic open baskets.

Procedures ii and iii were done in duplicate.

### Meat Yields

Four randomly chosen male crabs (average weight 2.56 lb.,±0.69 lb.) were steam cooked in a nonpressurized steam kettle for 20 minutes. Yields of body parts and picked meat were determined.

### Proximate Analysis

Percentages of protein, crude lipid and moisture in claws, body cores and legs were determined for raw and cooked crab meat using basic A.O.A.C. (1) methods for nitrogen, Bligh and Dyer (2) for total lipids, and moisture at 100°C for 24 hours in vacuum (15 inch, Hg). Determinations were carried out on four cooked and four raw male crabs. Duplicate samples from each crab were analyzed for

protein and moisture and single samples were analyzed for crude lipid.

Discoloration Inhibition

Live crabs were butchered to halves (a body core with four legs and one claw attached). In a plastic can, 50 gallons of 0.25, 0.50 and 1.00% sodium bisulfite solution (sodium metabisulfite, Virginia Chemical Co., Portsmouth, VA.) was prepared using sea water. The crab halves were then exposed to the bisulfite solutions as follows:

(i) Control 10 male and 10 female halves were separately packed in plastic bags and iced.

(ii) 0.25% 10 male halves were dipped for 20 seconds, in the 0.25% solution, bagged and iced.

(iii) 0.50% 10 male halves were dipped for 20 seconds, in the 0.50% solution, bagged and iced.

(iv) 1.00% 10 male and 10 female halves were separately dipped for 20 seconds in the 1.00% solution, bagged and iced.

Crab halves were monitored every 24 hours and the number of halves showing blackening recorded. Halves with more than 5% of the flesh surface discolored were judged to be unacceptable to consumers.

## RESULTS AND DISCUSSION

Because the Gulf of Mexico deep-sea fishing grounds can range in excess of 20 to 25 hours from port, and complete fishing trips can last beyond one or two weeks, proper methods of storage are necessary to preserve the crab quality during transport. Results from survival studies indicate that live on-board storage of deep-sea crabs is not recommended using the simple, economic storage methods (Table 1). Direct icing and open basket storage had mortality rates, for male crabs, ranging from 64 to 100% after three days. Burlap bag storage had a male crab mortality rate of 28% after two days. Female crabs appeared to be more durable than their male counterparts, but the mortality rates for all storage methods were still unacceptably high. Without increasing the sophistication of the storage method, it appears that butchering the crabs at sea and storing the parts on ice or cooked and frozen, may be required.

Deep-sea crabs are simply butchered by splitting them in half, removing the carapace and eviscerating. Butchering can be done by hand, aided with large knives and metal blocks designed with a fulcrum for splitting the carapace. Running water to help remove viscera and a brief drain time to allow bleeding from the parts is recommended. Automation is also available as used to butcher red

Table 1. Percent mortality of Geryon crabs harvested from the Gulf of Mexico and stored under refrigeration (50°F) in three separate conditions.

| | | PERCENT MORTALITY | | | | | |
|---|---|---|---|---|---|---|---|
| | | Open Basket | | Burlap Bag | | Iced | |
| Hours | Test | M | F | M | F | M | F |
| 24 | (A) | 72 | 40 | | | | |
| | (B) | 100 | 48 | | | | |
| 48 | (A) | | | 36 | 32 | | |
| | (B) | | | 28 | 12 | | |
| 72 | (A) | | | | | 64 | 11 |
| 96 | | | | | | | |
| 102 | (A) | | | | | 91 | 56 |

M - males
F - females

crabs aboard New England vessels. Butchered crabs yield two halves, each consisting of a body core with four legs and one claw attached. Butchered male red crabs are reported to yield 58.3% raw halves (4) and Gulf of Mexico crabs provide a similar yeild. If the claws are detached from the halves, the core-leg combination remaining is termed a cluster.

Whole, commercial size male crabs from the Gulf steamed 20 minutes yielded 84.7% of the precooked weight. The whole cooked crab includes 58% halves (cluster + legs + core), and when the halves are carefully picked they yield 22.9% (+2.1%) meat (Figure 1). Meat yields under commercial conditions will certainly be lower. Holmsen (5) found male crabs to yield 24% cooked meat under laboratory conditions but only 11% when the red crabs were picked in a commercial blue crab processing facility. Meade and Gray (7) found yields to increase to 20-22% after pickers were given a short training session. Typically, the red crab industry expects a 20% meat yield utilizing automated roller-type picking machines (High Seas Corporation and Bay Trading Co. Inc., Mass. personal communications).

A proximate analysis of the raw and cooked deep-sea crab flesh shows it to be a high protein low fat seafood (Table 2). The cooking technique increased the percent edible protein as a consequence of moisture loss.

Melanosis, the post mortem formation of a dark discoloration on the shell or flesh of crustaceans, has been well documented in shrimp. The discoloration is caused, in part, by a polyphenol oxidase enzyme that catalyzes the oxidation of phenols. The oxidized phenols undergo a series of reactions and eventually condense to form a dark melanin pigment. Constantinides and Chang (3) found G. quinquedens to contain multiple forms of polyphenol oxidase. They also noted that a short incubation period was necessary to activate the enzyme.

A black discoloration, believed to be melanosis, began to develop on iced butchered deep-sea crabs after about 24 hours. The discoloration began on any gill pieces not removed during butchering and on any flesh surfaces exposed to air. At later stages the shell became a grey-brown and then a black color. Some success in inhibiting the discoloration was achieved with sodium bisulfite dips (Table 3). A 1.00% dip afforded the male crab halves four days of protection against the discoloration. These preliminary results suggest melanosis may be more severe in the southern variety of Geryon harvested in warmer climates.

## CONCLUSIONS

Butchering crabs at sea and storing the butchered halves on ice appears to be a desirable alternative to live crab storage. The three simple economic methods of storage investigate all had unacceptably high mortality rates. It is felt that more sophisticated storage methods could be developed, but that the costs involved with these methods would be prohibitive.

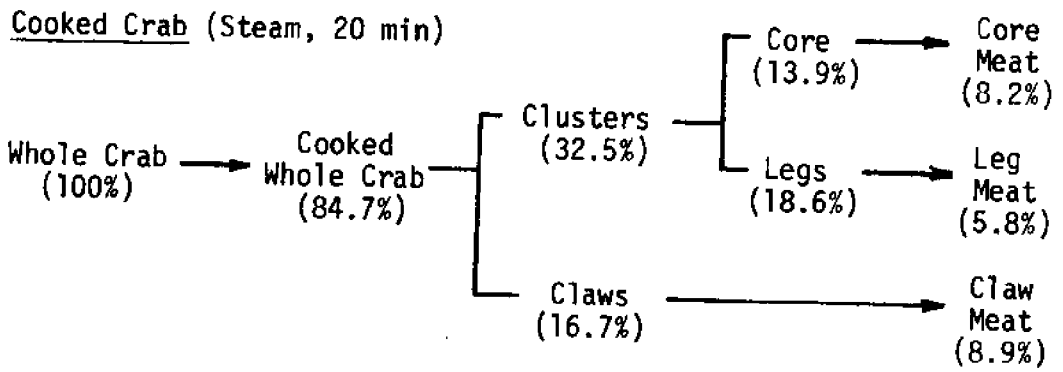



Figure 1. Edible yields from commercial size male Geryon crab harvested from the Gulf of Mexico. Yields are expressed as a percent of the original whole, raw crab weight.

Table 2. Basic chemical composition (percentage) from raw and cooked male Geryon crab harvested from the Gulf of Mexico.

Cooked Crab Meat

| | Leg | Core | Claw | Weighed Avg. |
|---|---|---|---|---|
| Protein (N x 6.25) | 18.32 | 19.15 | 19.22 | 18.99 |
| Crude Lipid | 1.4 | 2.8 | 1.5 | 1.9 |
| Moisture | 81.48 | 80.91 | 81.21 | 81.17 |

Raw Crab Meat

| | Leg | Core | Claw | Weighed Avg. |
|---|---|---|---|---|
| Protein (N x6.25) | 15.48 | 17.05 | 13.70 | 15.41 |
| Crude Lipid | 1.3 | 1.6 | 1.0 | 1.3 |
| Moisture | 84.18 | 83.54 | 84.20 | 84.13 |

Table 3. Percent of crab clusters which develop black discoloration such that the raw product would be objectionable to the consumer. All clusters were packed in plastic bags stored in refrigeration (35°F).

| Days Storage (35 F) | Sodium Bisulfite Dip (20 sec) | | | | | |
|---|---|---|---|---|---|---|
| | No Dip | | .25% | .50% | 1.0% | |
| | F | M | M | M | F | M |
| 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 1.1 | 0 | 0 | 0 | 0 | 0 | 0 |
| 2.1 | 50 | 80 | 10 | 0 | 0 | 0 |
| 3.0 | 70 | 90 | 10 | 0 | 0 | 0 |
| 4.2 | 70 | 90 | 10 | 0 | 0 | 0 |
| 5.2 | 100 | 100 | 50 | 10 | 10 | 60 |
| 6.1 | 100 | 100 | 60 | 20 | 30 | 60 |
| 7.1 | 100 | 100 | 100 | 20 | 50 | 80 |

M - males
F - females

Butchered male crabs were found to yield 22.9% (±2.1%) total meat when the steamed body parts were picked. Chemical analysis shows the meat to be a high protein low fat seafood.

Some success was encountered in controlling melanosis with sodium bisulfite dips. A 1.00% dip afforded deep-sea crabs with four days of protection against the discoloration. Before the potential of this fishery can be realized the melanosis must be controlled for significantly longer periods. New dip formulations and exposure times are currently being investigated. Further investigations will include the consequences of various cooking methods.

## REFERENCES


1. A.O.A.C. 1980. "Official Methods of Analysis," 12th ed. Association of Official Analytical Chemists. Washington, D.C.

2. Bligh, E.G. and W.J. Dyer. 1959. A rapid method of total lipid extraction and purification. Can. J. Biochem. Physiol. 37:912.

3. Constantinites, S.M. and G. Chang. 1976. Enzymatic blackening in the red crab. (Abst.) Annual Atlantic Fisheries Tech. Conf., Newport, RI.

4. Gerrior, P. 1981. Distribution and effects of fishing on the deep sea red crab, Geryon quinquedens, of southern New England. Masters thesis, Southeastern Massachusetts University, North Dartmouth, MA.

5. Holmsen, A.A. 1968. The commercial potential of the deep sea red crab (a progress report). University of Rhode Island, Department of Food and Resource Economics, Occasional Paper G8-138, April.

6. Holmsen, A.A. and H. McAllister. 1974. Technological and economic aspects of red crab harvesting and processing. University of Rhode Island, Marine Technical Report Number 28.

7. Meade, T.L. and G.W. Gray. 1973. The red crab. University of Rhode Island Marine Technical Report Series Number 11.

8. NMFS. 1982. National Marine Fisheries Service, Southeastern Fisheries Center, Pascagoula, MS. Computer print-outs documenting results from Oregon II Cruises for exploratory fishing about the Gulf of Mexico, 1956 thru 1976.

9. Sweat, D.E. and W.S. Otwell. 1983. Experimental gear for harvesting deep-sea crab (Geryon sp.) from the Gulf of Mexico. Proc. Eighth Annual Trop. and Subtrop. Fish. Tech. Conf., Tampa, FL.

# EXPERIMENTAL GEAR FOR HARVESTING DEEP-SEA CRAB (GERYON SP.) FROM THE GULF OF MEXICO

Donald E. Sweat
Marine Advisory Program
Largo, Florida 33540

and

W. Steven Otwell
Department of Food Science and Human Nutrition
University of Florida
Gainesville, Florida 32611

## INTRODUCTION

Exploratory fishing ventures during 1981 and 1982 by commercial boats operating in deep waters along the southwest coast of Florida have found a potential commercial resource of deep-sea crabs. Their results are limited, but have stirred interest amongst area fishermen, including traditional snapper-grouper vessels which are looking for additional and alternative sources of valuable production. These results coincide with the current decreasing and limited supply of traditional deep-sea crabs, king and snow crab. Regional processors have advocated a strong interest for additional crab resources which could be directed to the increasing market demand for whole crab parts and shucked crab meat. Thus, this study was initiated to determine feasibility of commercial harvest of the deep-sea crab (Geryon sp.) in the Gulf of Mexico.

## GEAR

Initially tours of the existing red crab fishery in New England were arranged to observe harvest gear, traps and on-board handling techniques. A modified bottom long-line with crab traps was selected for further investigation (figure 1). Modifications from the red crab gear included use of 3/4 inch polyproylene buoyline and groundline, and metal wire traps as opposed to wooden traps used in New England. Subsequent experience indicated a second buoyline was optional depending on currents which could entangle gear.

Two cruises to demonstrate harvest technology have been completed; July 6-13 and September 1-8, 1982. Each cruise was arranged in cooperation with commercial fisherman, Mr. Orlen Oakleaf, captain of the Margueritte 'B' fishing out of John's Pass inlet. This 68 foot fishing vessel was initially equipped to fish bottom long-lines or swordfish and shark gear. The only on-board gear modification was the addition of a hydralic driven pot puller used to retrieve the crab gear. The mode of operation was to fish crab gear to suit project objectives and fish long-line gear for swordfish

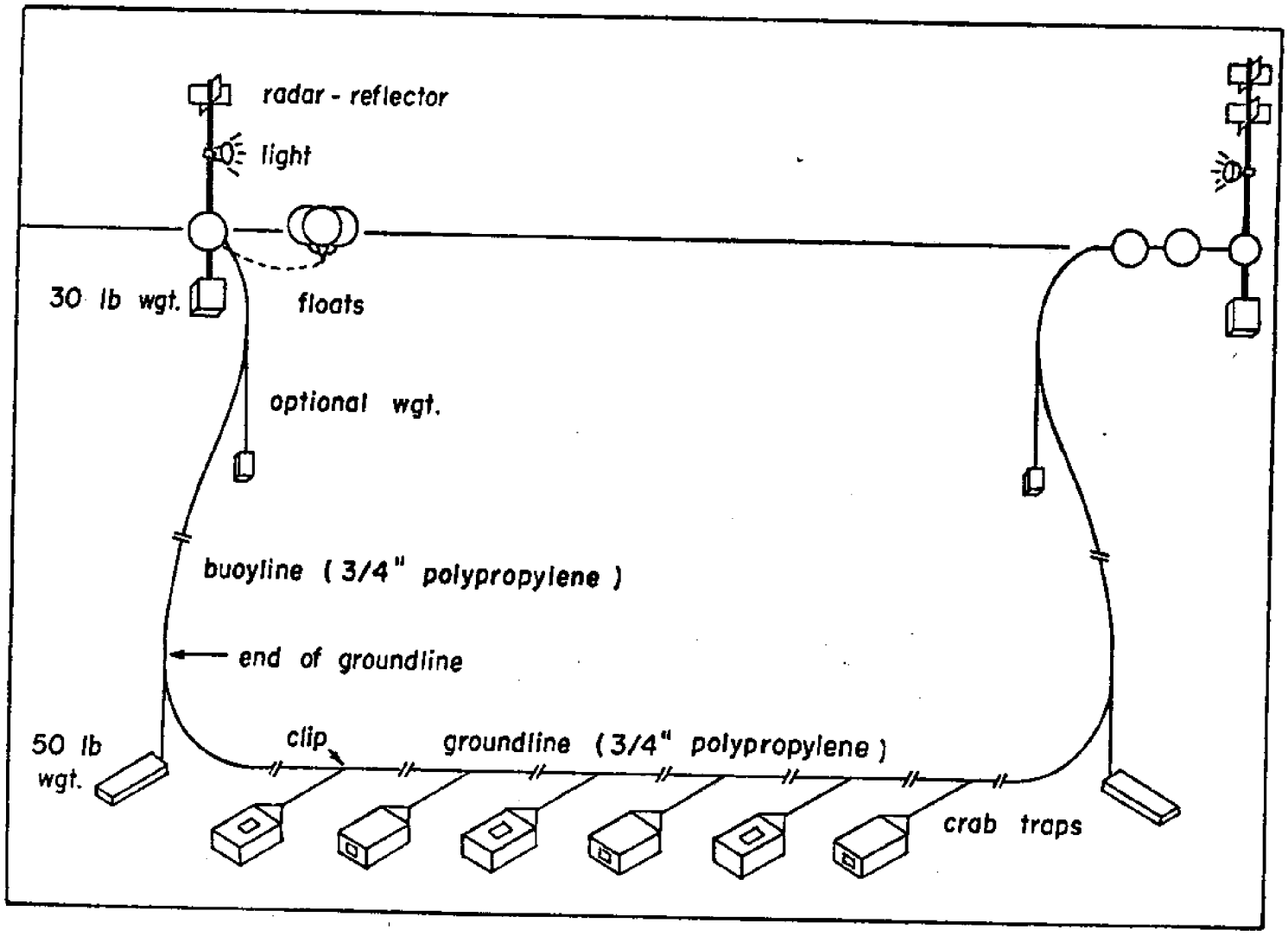



Figure 1. A 'set' of bottom long-line crab traps with two type traps (side or top entrance) and two optional arrangements for surface buoys and reflectors.

and shark as the work schedule permitted. The investigators obtained all harvest data including catch rates and monitoring crab size, bait performance, soak time, etc.

## FIRST CRUISE (July 6-13)

The objectives of the first cruise were to obtain experience with the crab fishing gear, check gear performance at various depths, use different baits, and note on-board handling problems. Two trap designs were used - collapsible and nested (figure 2). These traps were designed to conserve deck space. Six nested traps or twenty collapsible traps could be racked or collapsed to occupy the same space as two open traps of similar dimensions. One set implies one arrangement of gear (line, traps, etc.) placed in one location. Twenty traps were used per set, either alternating trap types or exclusive use of one trap type. Approximately 3 to 5 pounds of bait tied to net bags was used per trap. Various baits used included spanish sardines, pacific mackerel, tilefish or grouper heads. After baiting, one set could be lowered in place in approximately 30 minutes. Soaktime (duration on bottom) ranged from 13 to 22 hours.

Running time from port to the first fishing location was 18 hous. Eight sets were completed during this cruise. All sets were located within latitudes $27^0 10'$ and $26^0 50'$, and longitudes $84^0 50'$ and $85^0$. Fishing depths ranged from 210 to 340 fathoms. Surface water temperatures were between 85 to $87^0$F, bottom temperatures were not recorded. Substrata varied from a sand-mud mix to soft mud. Weather was hot ($>90^0$F), dry and calm. Two sets (310 and 340 fathoms) were nonproductive because they became fouled by excessive currents which rolled and tangled the gear.

Total production for the first cruise was 1,300 pounds of crabs, 750 pounds male and 550 pounds female. The nested traps were more productive than the collapsible traps (table 1). For the entire cruise including all nonfouled sets, 80 nested traps soaking 13 to 22 hours yielded 1,065 pounds of crab, 640 pounds male and 425 pounds female. Although the nested trap was heavier, the flimsy collapsible trap was more difficult to handle and the loose hung netting and insecure entrance may have deterred crabs. After the fourth set, use of the collapsible traps was discontinued. The collapsible trap was ideal for conserving deck space, but design improvements to secure netting and consider mesh size are recommended before further use.

There was no apparent pattern in total production by depth (210 to 310 fathoms) or soaktime (13 to 22 hours). Crabs were attracted to all baits, but two simultaneous sets using only nested traps, soaked for the same time in the same location (275 to 280 fathoms) caught over two times more crabs attracted to pacific mackerel than to grouper heads. The other sets utilizing spanish sardines and tilefish were placed in variable depths for various soaktimes which would influence interpretation of the results. Generally, the fresh, 'oily' baits (mackerel and sardine) were more successful and the firmer, longer lasting mackerel seems preferable.

Table 1. Catch rates for nested and collapsible traps used during the first cruise.

| Set | Soaktime (hrs.) | Trap (No.) | Crab/Trap | Males/Trap |
|---|---|---|---|---|
| 1 | 13 | Nested (10) | 15.9 | 3.4 |
| | | Collap.(10) | 3.7 | 1.0 |
| 2 | 14 | Nested (10) | 5.1 | 4.5 |
| | | Collap.(10) | 2.1 | 1.9 |
| 3 | 12 | Nested (10) | 5.2 | 1.5 |
| | | Collap.(10) | 1.8 | 0.2 |
| 4 | 14 | Nested (10) | 10.4 | 0.3 |
| | | Collap.(10) | 7.1 | 0.2 |
| 5 | 21 | Nested (20) | Fouled* | |
| 6 | 22 | Nested (20) | Fouled* | |
| 7 | 19 | Nested (20) | 9.2 | 5.3 |
| 8 | 17 | Nested (20) | 4.5 | 2.6 |
| Average | | Nested | 8.4 | |
| | | Collap. | 3.6 | |

*No productive set due to strong currents which tangled and fouled the gear.

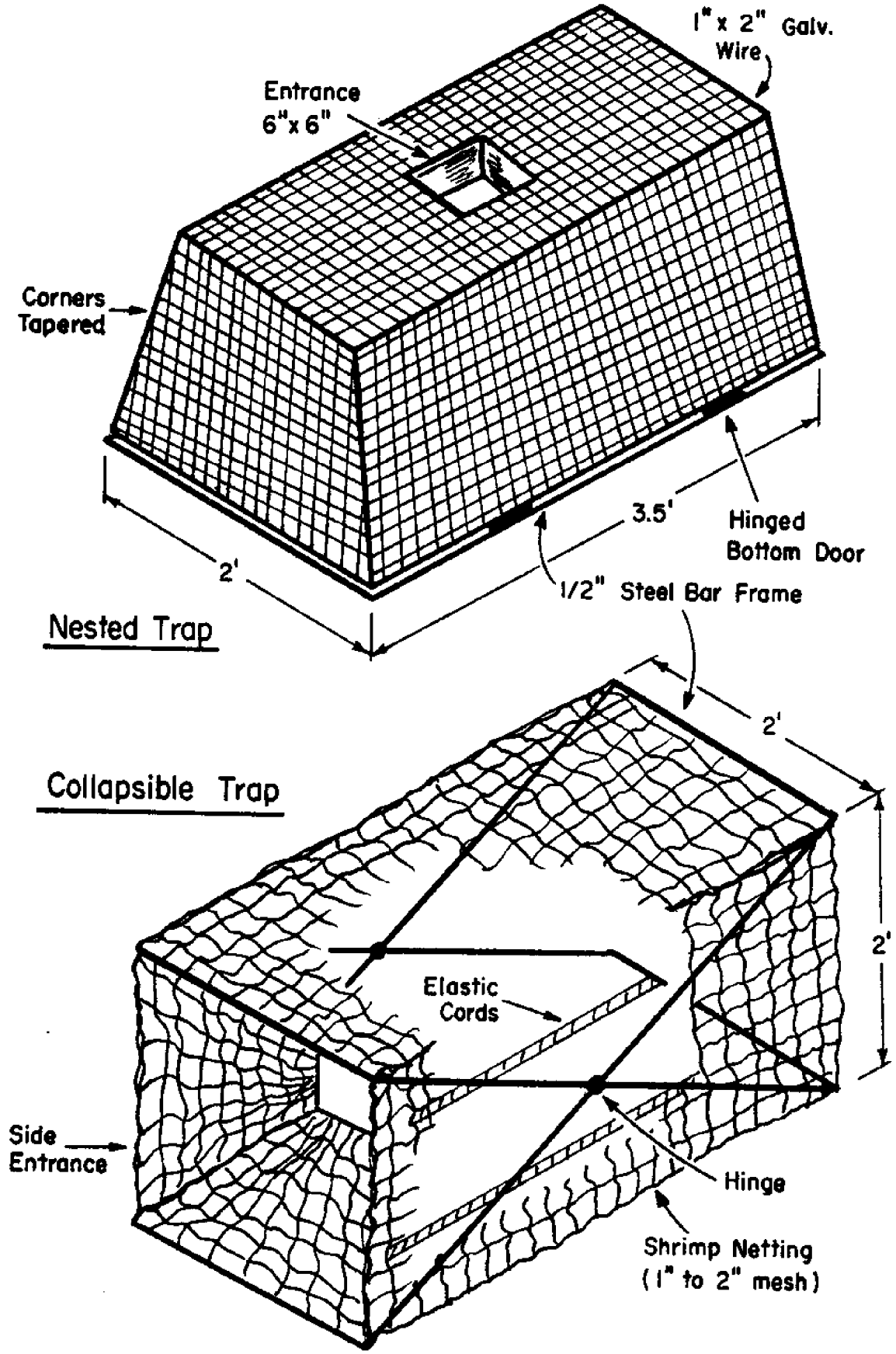



Figure 2. Nested and collapsible traps used during the first cruise.

As expected the harvest included both sexes and the larger male crabs were easily distinguished from the females. The average male was 6.2 ± 0.4 inches wide (carapace width) and weighed 2.5 ± 0.4 pounds. The average female was 4.9 ± 0.3 inches wide and weighed 1.1 ± 0.2 pounds. Size variations and ranges are listed in table 2. These sizes are substantially larger than the average commercial size red crabs harvested in New England. The average commercial (trap harvest) red crab is 5.0 inches wide and weighs 1.5 pounds, and the females, usually all less than 1.0 pound, are discarded. Surprisingly the average size females Geryon from the Gulf of Mexico were within the commercial size range for northern male red crabs.

Average male crab size was larger from the nested traps (table 2), but overall there was no significant difference in crab size for either sex relative to trap design, depth fished, soaktime or bait. Failure to harvest smaller crabs suggest distribution by size.

Sex ratio per trap design was similar, but more females were harvested at shallower depths (table 3). More males were harvested in depths greater that 275 fathoms. This distribution is similar as reported for red crabs harvested in New England.

Between crab sets, two evenings of surface long-lining (approx. 10 miles of gear) produced a supplementary catch of swordfish and sharks. Thus, two fishing activities could be conducted from the same vessel with the same crew.

## SECOND CRUISE (September 1-8)

Objectives of the second cruise were to use additional trap designs, variable soaktimes, and initiate more on-board handling and processing. The gear was similar as used on the first cruise except more traps were used per set and one buoyline was eliminated to prevent risk of tangling due to currents. Mackerel, spanish and pacific was the only bait. Isolated attempts to use catfood (16 oz. cans) attracted crabs but did not last.

Eleven sets were completed all within latitudes $27^0 45'$ and $28^0$, and longitudes $85^0 9'$ and $85^0 16'$. This location was north of the first cruise. Surface water temperatures were similar to the first cruise, but climatic conditions shifted to rainy weather with 10 to 20 mph winds and 30 mph gusts. The gear could be successfully fished during these conditions.

Depth fished was restricted from 245 to 290 fathoms to concentrate effort in depths previously noted to contain a higher proportion of males. Soaktimes were originially selected to vary from one to four days but an unexpected concentration of 'sea lice' (½ inch isopods, unidentified) preyed on the bait thus, restricted soaktime from 17 to 42 hours. In one particular set, sixty pounds of bait in the largest trap was devoured by isopods within 24 hours. Without bait the traps failed to attract crabs.

Table 2. Size crabs harvested with nested and collapsible traps during the first cruise.

| Trap | Sex | Carapace Width (inches) | | | Whole Weight (pounds) | | |
|---|---|---|---|---|---|---|---|
| | | Range | Mean | Std. dev. | Range | Mean | Std. dev. |
| Nested | Male | (5.5-7.0) | (6.2) | (0.4) | (1.6-3.7) | (2.5) | (0.4) |
| | Female | (3.9-5.7) | (4.9) | (0.3) | (0.7-1.6) | (1.1) | (0.2) |
| Collapsible | Male | (5.5-6.6) | (6.1) | (0.3) | (1.8-3.1) | (2.4) | (0.3) |
| | Female | (4.3-5.7) | (4.9) | (0.3) | (0.8-1.8) | (1.1) | (0.2) |

Table 3. Crabs harvested by depth, only using data from nested traps from the first cruise.

| Set | Depth (fathoms) | Total Crab Caught | Percent Males |
|---|---|---|---|
| 4 | 210 | 104 | 3 |
| 3 | 245 | 52 | 29 |
| 1 | 260 | 159 | 21 |
| 8 | 275 | 90 | 58 |
| 7 | 280 | 183 | 58 |
| 2 | 310 | 51 | 88 |

Traps included the original nested design, a new boxed design and one large square trap affectionately called the 'condo' (figure 3). The box design provided similar volume as in the nested trap but the box trap was easier to build. The box trap did not conserve deck space. Entrances on box traps were slightly smaller than on the nested traps. Some of the box traps had two end entrances, and others had one top entrance as on the nested trap.

For the entire cruise including eleven sets,253 traps caught over 1,950 pounds of crabs. Catch rates for nested and boxed traps were only recorded separately for the first 6 sets. Overall the nested traps were more productive and caught more males than the boxed traps (table 4). This difference in catch rate may be explained by the slightly larger entrance on nested traps. There was no significant difference in catch rate for box traps with end or top entrances. The condo was only set twice but harvested 143 crabs. Compared to the smaller traps fished on the same set, the condo caught as many crabs as 8 or 22 nested and boxed traps depending on location and soaktime. Likewise the condo caught a higher percentage of males. The better performance of the condo was attributed to the larger and tapered entrance.

There was a direct relationship between soaktime and total crabs caught in the smaller traps (table 4). Soaktimes greater than 24 hours were more productive. Soaktimes longer than 42 hours were not possible due to isopod predation on bait. Soaktimes for the condo were limited to only two sets, 20 and 42 hours. The longer condo set caught less crabs, but it can not be determined if the lower catch was due to location or possible escapement through the larger entrance after the bait desipated.

There was no pattern in total crabs, crab size or sex ratio caught per set across all depths (245 to 290 fathoms). Total catch depended on soaktime, and sex ratio was influenced by trap design. Overall the average male crab weighed 2.3 pounds and measured 6.2 inches (carapace width). The average female weighed 1.0 pounds and measured 4.9 inches. Variations and ranges in size are listed in table 5. There was no significant difference in crab size per trap designs. Although the average crab size was smaller from the second cruise, these crabs were substantially larger than the commercial New England red crabs and the average female crabs were just large enough to suit the New England commercial grade.

The sex ratio from depths of 245 to 290 fathoms was not as expected when compared to results from the first cruise. The smaller traps consistently caught a higher proportion of females from all depths (table 6), but the larger condo caught more males. Again the larger, tapered entrance may account for the performance of the condo. This is clear evidence that the sex ratio in the harvest can be influenced by trap design and may not reflect the actual ratio at particular depths. Soaktime did not influence the harvest sex ratio.

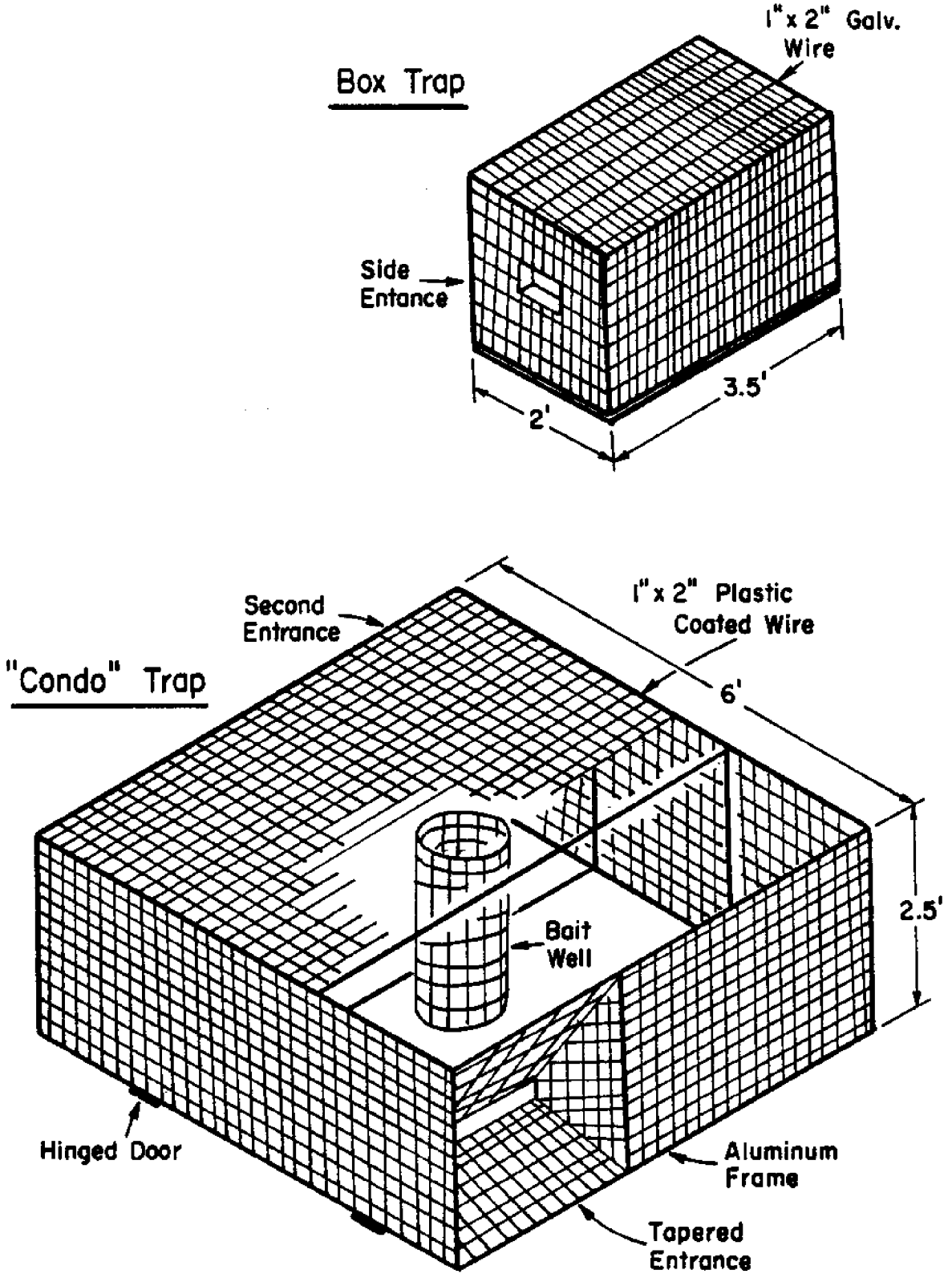



Figure 3. Additional trap designs used during the second cruise.

Table 4. Catch rate by soaktime for crabs caught during second cruise.

| Set | Soaktime (hrs.) | Trap (No.) | Crab/Trap | Males/Trap |
|---|---|---|---|---|
| 10* | 17 | Nested(12)<br>Boxed (12) | 1.7 | 0.3 |
| 11* | 18 | Nested(12)<br>Boxed (10) | 3.9 | 1.3 |
| 8* | 19 | Nested (6)<br>Boxed (18) | 4.8 | 0.3 |
| 9* | 20 | Nested<br>Boxed (19) | 3.8 | 0.6 |
| | | Condo (1) | 85.0 | 40.0 |
| 5 | 23 | Nested (5) | 5.0 | 2.4 |
| | | Boxed (17) | 3.0 | 0.8 |
| 7* | 24 | Nested(12)<br>Boxed (12) | 4.4 | 1.2 |
| 2 | 24 | Nested (6) | 8.8 | 2.3 |
| | | Boxed (18) | 3.6 | 0.9 |
| 6 | 26 | Nested(12) | 7.2 | 2.2 |
| | | Boxed (12) | 5.1 | 0.9 |
| 3 | 27 | Nested (5) | 7.8 | 2.8 |
| | | Boxed (17) | 6.7 | 1.9 |
| 1 | 40 | Nested(12) | 8.2 | 2.1 |
| | | Boxed (12) | 9.1 | 1.8 |
| 4 | 42 | Nested (5) | 7.4 | 2.4 |
| | | Boxed (16) | 8.0 | 2.2 |
| | | Condo (1) | 58.0 | 39.0 |

*For station 7-11 catch data was recorded pooled for nested and boxed traps.

Table 5. Size crabs harvested with nested, boxed and condo traps during second cruise.

| Trap | Sex | Carapace Width (inches) Range | Mean | Std. dev. | Whole Weight (pounds) Range | Mean | Std. dev. |
|---|---|---|---|---|---|---|---|
| *Nested and Boxed | Male | (5.6-7.3) | (6.2) | (0.3) | (1.3-3.5) | (2.4) | (0.4) |
| | Female | (4.4-5.8) | (4.9) | (0.3) | (0.6-1.5) | (1.0) | (0.2) |
| **Condo | Male | (5.6-7.1) | (6.3) | (0.3) | (1.7-3.1) | (2.4) | (0.3) |
| | Female | (4.2-5.7) | (5.0) | (0.3) | (0.8-1.5) | (1.1) | (0.2) |

*Size data recorded pooled for nested and boxed traps for stations 1-6.
**Size data collected for condo from stations 4 and 9.

Table 6. Data reflecting sex ratio for crabs harvested with nested, boxed and condo traps during the second cruise.

| Trap | No. Traps | Total Crab Caught | Percent Males |
|---|---|---|---|
| *Nested | 45 | 339 | 31 |
| *Boxed | 92 | 532 | 27 |
| **Condo | 2 | 143 | 55 |

*Data listed for nested and boxed traps from stations 1-6.
**Data listed for condo from stations 4 and 9.

Complimentary fishing activity included one evening of surface long-lining (12 miles) for swordfish.

With cooperation from the University of Florida's State Museum in Gainesville and the Florida Department of Natural Rsources in St. Petersburg, crab samples from our work have been mailed to the National Museum of Natural History, Smithsonian Institute. Dr. Raymond Manning, the Institute's Curator of Crustaceans has indicated these samples may represent an entirely separate form distinct from the red crab in New England. Further samples will be mailed to assist confirmation.

## SUMMARY

Data from the two cruises prove that large Geryon are present in all depths fished and they can be harvested with the described gear. Selection of trap design, bait, depths and soaktime will influence the catch. Interpretations from the harvest data are somewhat speculative realizing the limited number of gear sets and broad geographical range fished. Also non-uniform distribution and spotted congregation of the crabs would influence results. Contradictions in existing literature are a particular consequence of these factors. Thus, the results relative to crab distribution should be viewed as probable guides rather than final conclusions. These observations may change with further experience.

1. Crabs are present in depths from 200 to 300 fathoms with a higher proportion of males in deeper water.

2. Crabs are attracted to all fish baits but a firmer, high oil content bait is recommended. The bait must be in good condition, not spoiled. Five to ten pounds of bait is satisfactory for the small traps depending on soaktime and isopod predation. Traps with no bait do not catch crabs.

3. Nested traps with top or side entrances are recommended and allow stacking to save deck space. Entrance modifications and larger traps should be investigated.

4. Soaktime should exceed 24 hours. Longer soaktimes catch more crabs and allow complimentary fish activity.

# PURSE SEINE FISHING FOR COASTAL PELAGIC SPECIES IN THE NORTHERN GULF OF MEXICO

Charles M. Roithmayr
National Marine Fisheries Service
Mississippi Laboratories
Pascagoula, MS 39567

## INTRODUCTION

A review of available information indicates that several species such as thread herring, Spanish sardine, round scad, scaled sardine and round herring which are found in schools in the southeastern waters of the U.S. could be harvested at a higher level than at present. Potential yields are estimated for the herrings, sardine and scad to be over one million metric tons in the northern Gulf of Mexico. Little reliance can be placed in estimates of their potential for fishery development until more is known about current landings, species and size composition as well as catch effort data.

The Southeast Fisheries Center (SEFC) initiated a project in July 1981 to monitor the coastal pelagic purse seine fishery of the northeastern Gulf of Mexico. The objectives are 1. to determine the species and size composition of commercial purse seine catches in the southeastern U.S.; 2. to collect data on total vessel catch and effort; and 3. to measure environmental parameters associated with fish catches, i.e., surface temperature, surface salinity and water turbidity. Data were collected by observers who boarded purse seine vessels and sampled the catch and recorded catch and effort data.

## METHODS

### Fishing vessels and gear.

Single boat purse seine vessels using hydraulic power blocks are 45 to 65' long. The size of the seine is approximately 275 fm. long and 16 fm. deep, and the mesh size is usually 1". The purse seine, Figure 1, is fished at the surface with the upper edge of webbing held by the corkline. The bottom edge of the seine is held down by the leadline. Attached by bridles is a series of rings through which the purse line is threaded. The purse line closes the bottom of the seine when a haul begins, trapping the fish that have been surrounded. The fishermen often rely on a spotter pilot to locate schools and to direct the boats to surround the school.

The net is hauled by a hydraulic powered power block, Figure 2. A large aluminum sheave supported by a rigid aluminum frame with

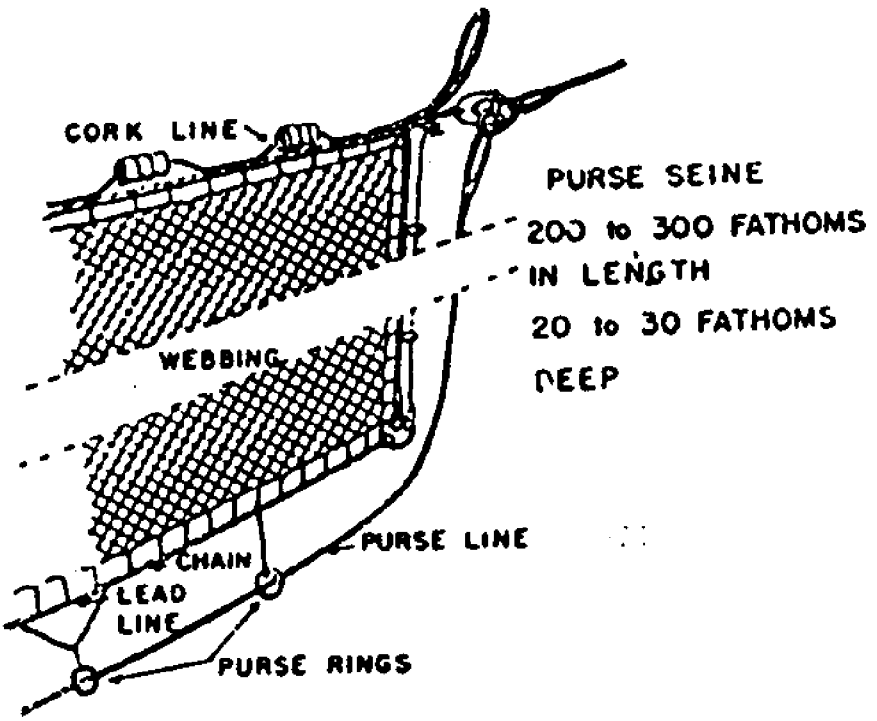



FIGURE 1. DIAGRAM OF A PURSE SEINE.

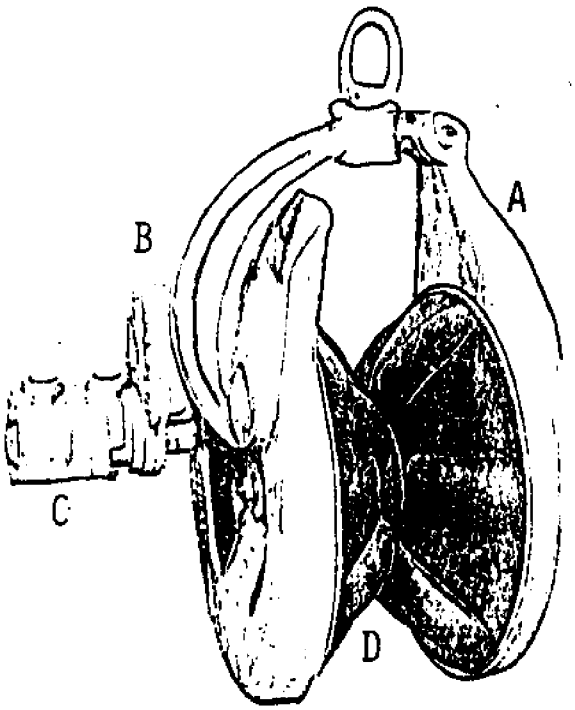



FIGURE 2. COMPONENTS OF THE PURETIC HYDRAULIC POWER BLOCK. A. ALUMINUM HOUSING. B. HINGED SIDE OF BLOCK. C. HYDRAULIC HOSE CONNECTION. D. ROTATING V-SHEAVE WITH NON-SKID RUBBER FACING AND CLEATS.

non-skid rubber facing is driven by a medium pressure hydraulic unit. It weighs less than 200 lbs. and is capable of exerting line pulls up to 1½ tons. A crew of 5 to 6 is usually required to purse the net. Figure 3 shows the sequence of the purse seine operation once the fish school has been detected from the vessel or by the spotter pilot. First the school is surrounded when the seiner runs in a circle setting the net and returning to the skiff which is attached to one end of the net. Second, a large weight called the "tom" is dropped overboard to insure that the net stays on the bottom during pursing. Third--the purse rings and leadline are hauled in by the power block. The fish are loaded aboard by means of a large brail or dip net.

Recently a new combination fishing vessel, the *Fisherman's Pride*, was built by Raffield Fisheries at Port St. Joe, FL which has the ability to purse seine in deeper water to 35 fm. The vessel is 62' long and 24' wide. She is powered by a 365 HP, 8 cylinder Caterpillar engine. Cruising speed is 10 knots at 1,800 RPM, and burns 16 gallons of fuel per hour. This is primarily intended for single boat purse seining with a power block and can be converted to trawling and longlining for bottomfish and reef fish. The vessel has a capacity of 50 tons and is equipped with refrigerated recirculating seawater system to preserve the quality of fish.

The fishing grounds for the single boat purse seine fleet include coastal waters, 2 to 10 fm., between the Mississippi River Delta and Pascagoula, MS; the Florida panhandle between Pensacola and Apalachicola, FL; and the west-central Florida coast between St. Petersburg and Sarasota, Figure 4. The principal ports of landing include Ysckloskey, LA; Pascagoula, MS; Destin, Panama City, Port St. Joe, and Cortez, all in Florida. Charlotte Harbor near Fort Myers, FL was once a major port when a reduction plant processed thread herring for fish meal; but legislation prohibiting purse seines in Florida state waters along the lower Gulf coast ended the fishery.

Purse seine fishing using the power block has several drawbacks insofar as the speed with which the fish can be captured and the quality of fish produced. It is slow because the seine must pass through the block and be manually stacked on the deck. During the time that the fish are hardened up, and while still confined in the net, fish quality decreases because the fish are damaged from abrasion and scale loss.

The drum seine has proven to be much superior to the power block in California and Washington because recovery of the net is nearly twice as fast as the power block. Not only does quicker retrieval result in less fish damage but it also results in shorter trips, thereby making drum seining more efficient and profitable. The system, Figure 5, consists of a hydraulically driven drum that extends across the stern of the vessel. The drum rotates both forward and reverse for setting and retrieving the net. The net is retrieved over the stern roller through a level wind which moves

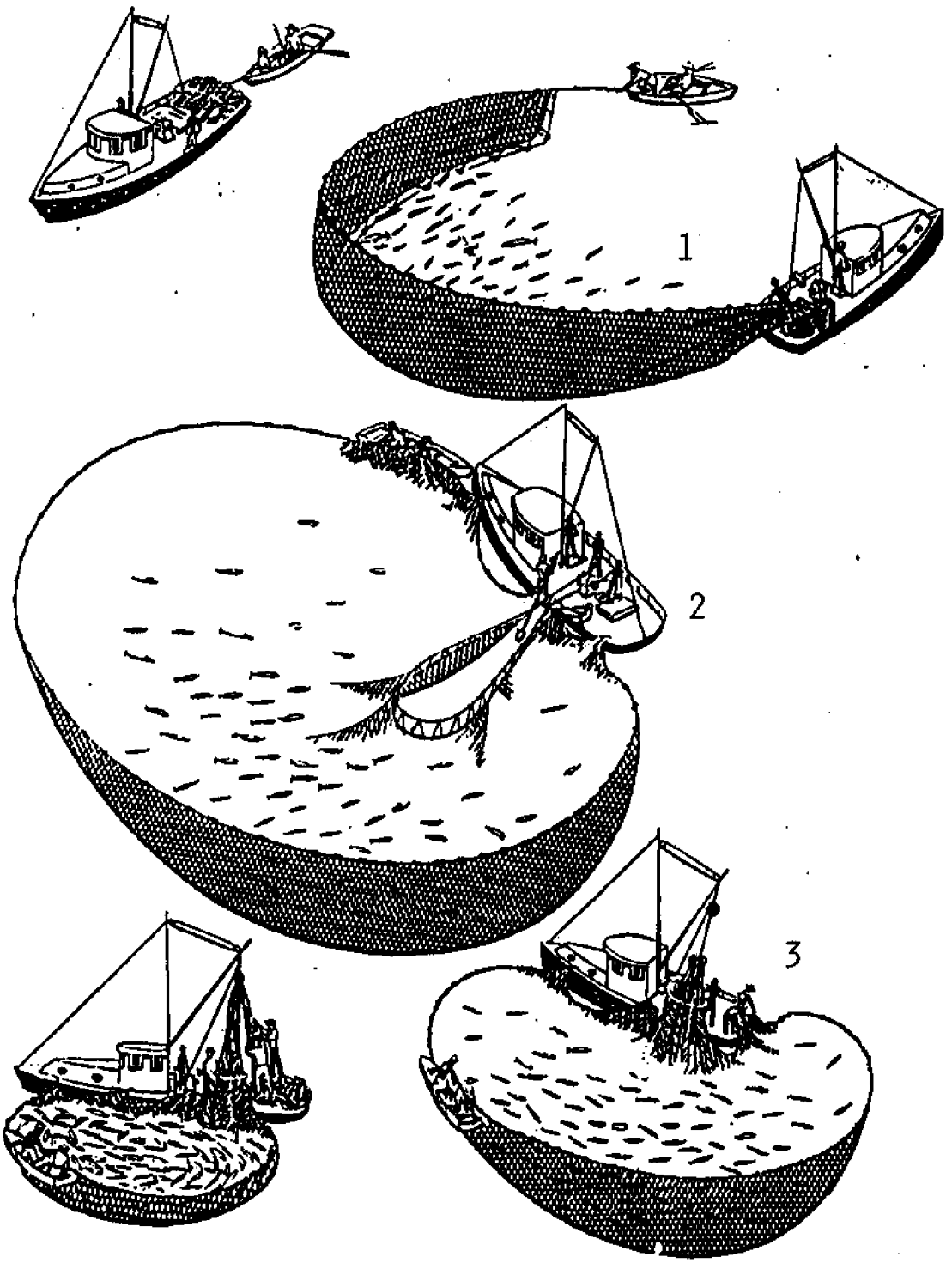



FIGURE 3. SEQUENCE OF PURSE SEINE OPERATION.
1. SETTING THE NET. 2. PURSING.
3. HAUL IN PURSE RINGS AND LEADLINE.

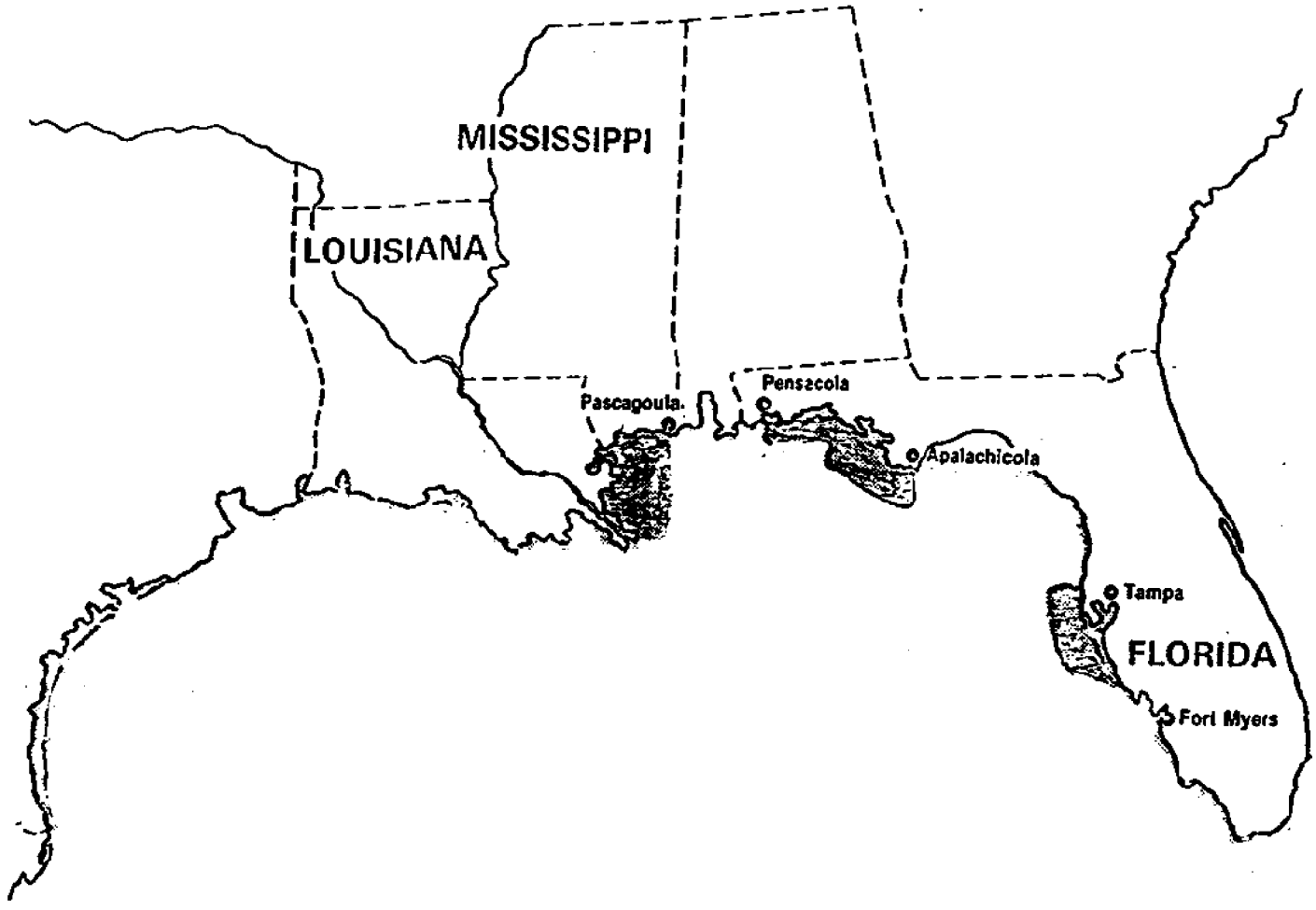



FIGURE 4. COMMERCIAL PURSE SEINE GROUNDS FOR COASTAL PELAGIC SPECIES.

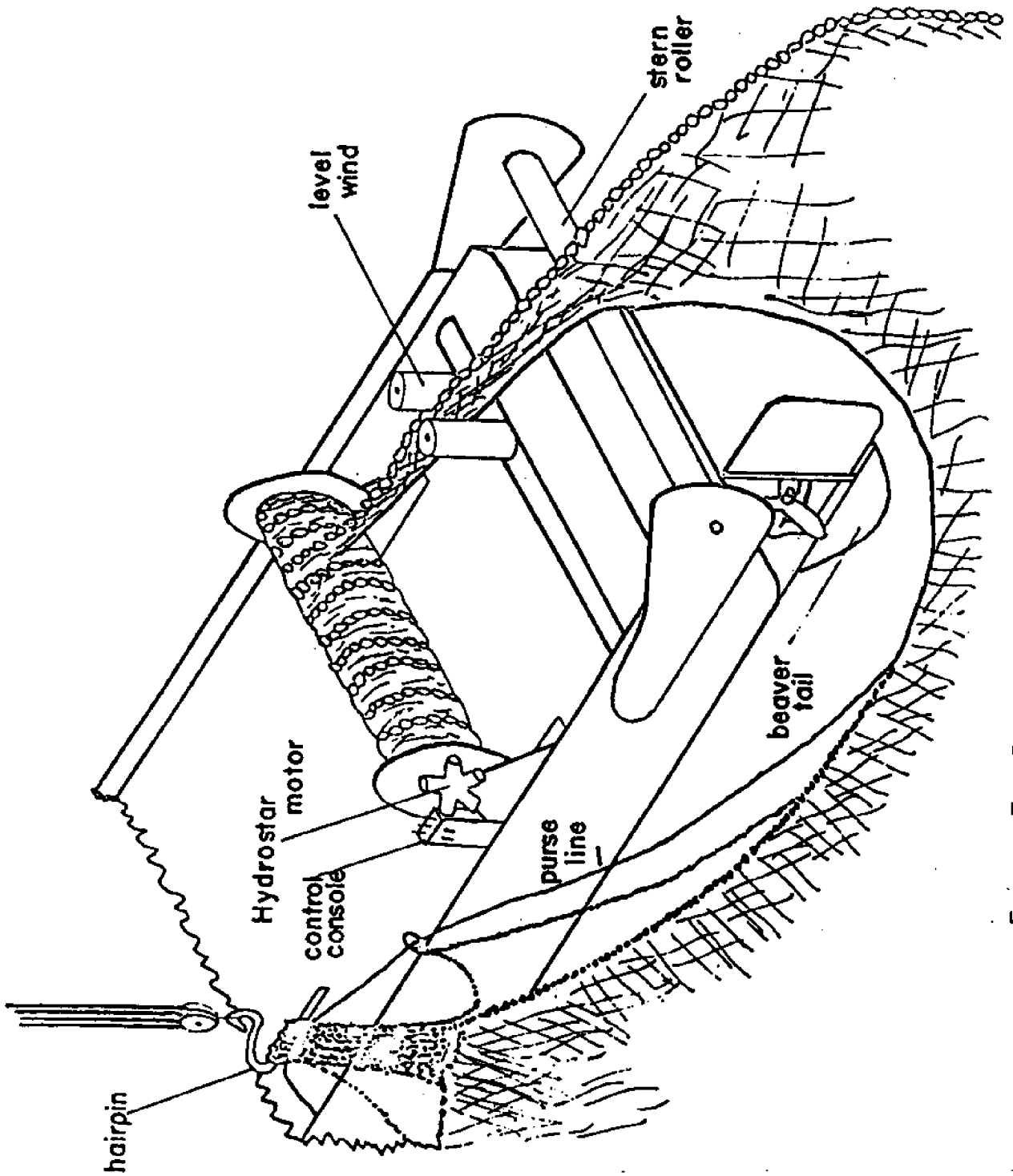



FIGURE 5. DIAGRAMMATIC LAYOUT OF A DRUM SEINE INSTALLATION.

back and forth across the deck on a track. The level wind consists of two parallel upright rollers that are tipped down when the net is unreeled for the setting. A freewheeling device allows the drum to run free when the net is being set and a brake controls the drum speed to prevent backlash. The hairpin is a steel rod which holds all the purse rings. It allows the purse line to slip freely through the rings while dropping off each ring as the net is reeled onto the drum.

Setting the drum seine consists of encircling the school by unreeling the net from the drum while the seine skiff is underway at 2 to 3 knots, Figure 6. The net is attached to the motorized skiff to provide the initial drag which pulls the net off the drum. Setting is complete when the seiner meets the skiff and the purse lines and tow lines are attached to the mid-ship davit.

Pursing the net is accomplished when the purse line is taken in by the winch and coiled on deck, Figure 7. Pursing is complete when the purse rings surface and are slipped onto the hairpin.

Hauling in the net occurs when the engineer reels in the net with the corkline over the stern roller on the drum, Figure 8. The level wind distributes the net evenly across the drum face as it winds on the drum. At the same time the purse rings slip off the hairpin and are rolled up with the net together with the purse lines.

## RESULTS AND DISCUSSION

### Single boat purse seine with power block.

A summary of the activity for the coastal herrings project for the 2-year period is given in Table 1. Ninety-five samples were collected during the 5-month period, July through November, 1981. Nine species were represented including mullet, Spanish sardine, round scad, Spanish mackerel, menhaden, thread herring, ladyfish, bluerunner and little tuna. The total sample weight amounted to 4,600 lbs. with a total live catch sampled amounting to 1,801,000 lbs. In 1982, 149 samples were drawn during the 7-month period, June through December. Eleven species were present including mullet, jack crevalle, bluerunner, menhaden, ladyfish, scaled sardine, Spanish sardine, round scad, bumper, bluefish and little tuna. Total sample weight amounted to 5,500 lbs. with the total live catch sampled being 1,217,000 lbs.

The catch effort data for the principal species caught by the purse seine vessels which were sampled by observers in Florida and Louisiana during the 1981 fishing season is given in Table 2. Thread herring yielded the highest average catch per set of 18 tons; followed by menhaden, 15 tons; ladyfish, 11 tons; round scad, 10 tons and mullet, 4 tons.

The average size composition data of the target species, Table 3, varies from 13 cm for round scad to 51 cm for ladyfish. Intermediate sizes consist of 17 cm for thread herring, 19 cm for menhaden and 34 cm for mullet.

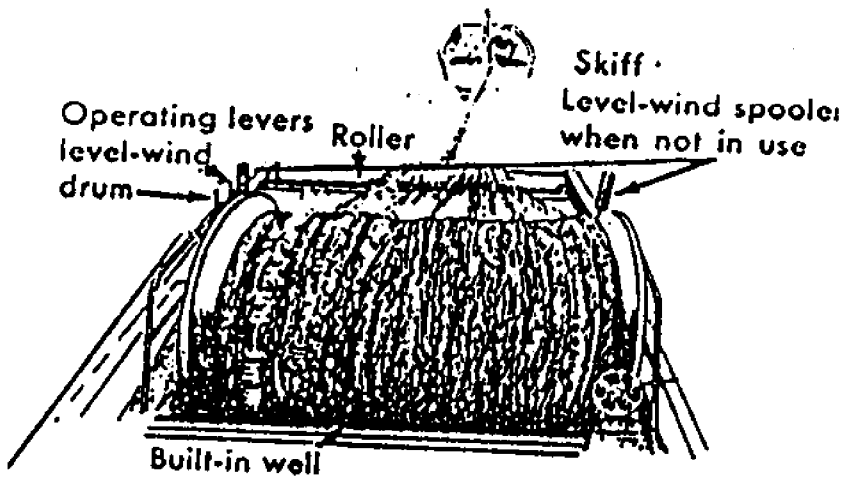



FIGURE 6. STARTING A DRUM SEINE SET.

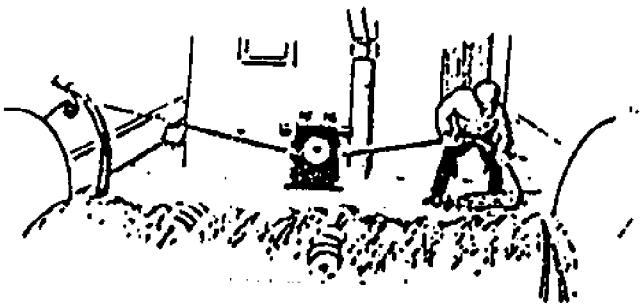


FIGURE 7. PURSE LINE IS TAKEN IN BY WINCH TO PURSE THE DRUM SEINE.

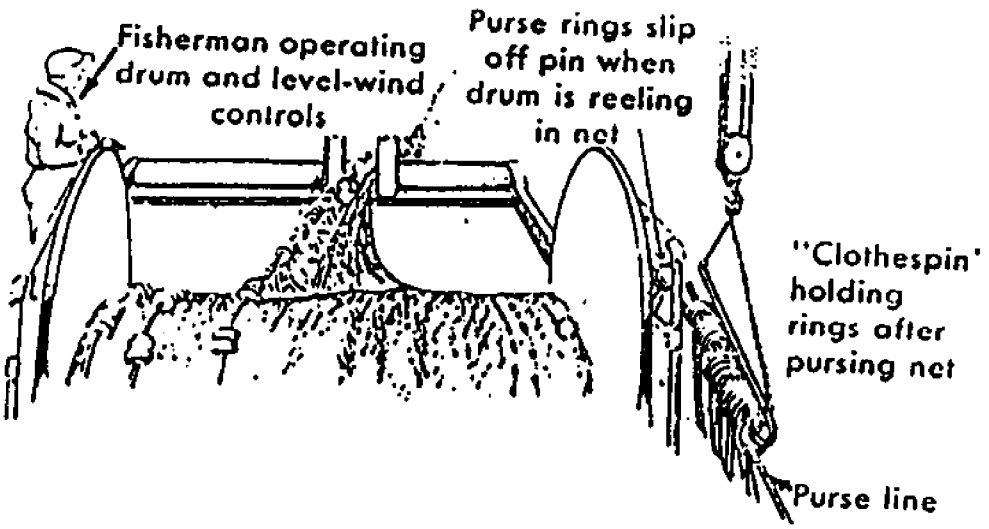



FIGURE 8. HAULING IN THE DRUM SEINE. HYDRAULIC SYSTEM OPERATES THE DRUM.

TABLE 1. ACTIVITY SUMMARY - SEFC COASTAL HERRINGS PROJECT, 1981-82

| | 1981 JULY-NOV. | 1982 JUN.-DEC. |
|---|---|---|
| TOTAL LIVE CATCH SAMPLED, THOUS. LBS. | 1,801 | 1,217 |
| NO. DAYS FISHED | 69 | 81 |
| NO. SPECIES SAMPLES | 9[1/] | 11[1/] |
| NO. SAMPLES[2/] | 95 | 149 |
| TOTAL SAMPLE WT., THOUS. LBS. | 4.6 | 5.5 |

1/ SPECIES INCLUDES JACK CREVALLE, BLUERUNNER, MENHADEN, LADYFISH, SCALED SARDINE, SPANISH SARDINE, ROUND SCAD, BUMPER, BLUEFISH, LITTLE TUNA, THREAD HERRING, SPOT.

2/ SPECIES COMPOSITION AND SIZE COMPOSITION.

TABLE 2. CATCH EFFORT DATA FOR SAMPLED PURSE SEINE VESSELS, FLORIDA & LOUISIANA, 1981

| SPECIES | AVERAGE CATCH/SET TONS | CATCH/SET RANGE-TONS | NO. SETS |
|---|---|---|---|
| THREAD HERRING | 18 | 6-53 | 8 |
| MENHADEN | 15 | 5-25 | 11 |
| LADYFISH | 11 | 2-20 | 15 |
| ROUND SCAD | 10 | 2-22 | 5 |
| MULLET | 4 | 1-8 | 6 |

TABLE 3. SIZE COMPOSITION DATA FOR PURSE SEINE SPECIES, FLORIDA & LOUISIANA, 1981

| SPECIES | AVERAGE FORK LENGTH CM | SIZE RANGE CM | NO. |
|---|---|---|---|
| THREAD HERRING | 17 | 10-21 | 828 |
| MENHADEN | 19 | 10-23 | 1,336 |
| LADYFISH | 28,51 | 23-56 | 1,248 |
| ROUND SCAD | 13 | 9-18 | 511 |
| MULLET | 34 | 19-39 | 743 |

The species composition for Florida purse seine catches sampled during October 1981 is given in Table 4. The total catch amounted to 299 tons of which 41 percent was thread herring, 19 percent ladyfish, 16 percent spot, 15 percent menhaden and 9 percent Spanish sardine and round scad.

Early results of the fishing operations by the new purse seine vessel *Fisherman's Pride* indicate the deepwater purse seine is working well with new strategies reflecting the use of the vessel as a mother ship for the catcher fleet. Over 250 tons of Spanish sardines, little tuna, menhaden, mullet and ladyfish were harvested during the first 5 days of fishing. By catching a variety of species that are available to purse seiners both nearshore and offshore, more of the production potential of underutilized or surplus stocks is being realized.

Single boat purse seine with power drum.

Anderson Seafoods, Inc. of Panama City, FL and Clark Seafood Co. of Pascagoula, MS were funded by the Gulf and South Atlantic Fisheries Development Foundation under a NMFS Cooperative Agreement to determine whether drum seining for underutilized species is feasible in the northern Gulf of Mexico. Preliminary results of the drum seine demonstrations were published in a final report by Harrell, 1981.

Anderson converted a steel hull shrimp trawler with a hold capacity of 75 tons. Seven trips, 5 to 10 days each, produced 359 tons of underutilized species. Seventy-five percent of the total catch consisted of bluerunner and redfish. Included in the catch was mullet, little tuna, butterfish, black drum and spadefish. The average daily catch amounted to 3.5 to 7.0 tons depending on the total number of fishing days which was not reported. The operation was marginally profitable because of high operational costs associated with increased fuel consumption and labor. The higher costs were incurred by the 2 large carrier boats used to transport fish to the dock. Only 1 small seine skiff was needed to assist in the capture of fish schools. The 200-mile trip from the fishing grounds off Louisiana and Mississippi to the home port of Panama City also increased the fuel costs. The vessel sonar located fewer schools than did the aircraft spotter pilot employed by Clark.

Clark constructed a new seine vessel with a capacity of 80 tons. Seven trips, 1 to 2 days each yielded 149 tons. Ninety-eight percent of the total catch consisted of little tuna, croaker, bluerunner and redfish. Other species included butterfish, jack crevalle, lookdown and shark. The average daily catch for 9 days' fishing was 16.5 tons. Clark's operation was more profitable than Anderson's because only 1 auxiliary vessel was used; the spotter located more schools than did the vessel sonar; and Pascagoula is situated close to the fishing grounds.

Following the demonstrations, commercial catch data were obtained by a NMFS observer aboard a drum seiner owned by Clark

TABLE 4. SPECIES COMPOSITION OF PURSE SEINE CATCHES, FLORIDA, OCTOBER, 1981

| SPECIES | TONS | PERCENT COMPOSITION |
|---|---|---|
| THREAD HERRING | 121 | 41 |
| LADYFISH | 57 | 19 |
| SPOT | 48 | 16 |
| MENHADEN | 46 | 15 |
| SPANISH SARDINE & ROUND SCAD | 27 | 9 |
| TOTAL | 299 | |

Seafood Co. in August 1982. The catch data are summarized in Table 5. Sixty-nine tons were caught in 6 sets during 2 days of fishing in 8 to 10 fm. southeast of the Mississippi River Delta. The average catch per set was 11.5 tons. Seven species were caught including croaker, sand seatrout, silvereel, bluerunner, red drum, shark and Spanish mackerel.

In early December 1982 a NMFS observer reported 5 sets were made on roe mullet by a Pascagoula based drum seine vessel in Mississippi Sound. The fish were lost on 1 set because of the seine fouling on bottom debris. The remaining 4 sets yielded 10 to 40 tons each.

REFERENCE

Harrell, Mona R. 1981. Results of exploratory drum seining efforts. Final report. Gulf and South Atlantic Fisheries Development Foundation, 46 pp.

TABLE 5. SUMMARY OF CATCH DATA BY R AND R, COMMERCIAL DRUM SEINER, AUGUST, 1982

| | |
|---|---|
| No. of Days | 2 |
| No. of Sets | 6 |
| Net Size | 350 fm X 30 fm |
| Crew Size | 6 |
| Location | SE Mississippi Delta |
| Depth | 8-10 fm |
| Total Catch | 69 tons |
| Avg. Catch/Set | 11.5 tons[1/] |
| No. Species | 7 |
| Species | Croaker[2/]<br>Sand Seatrout[2/]<br>Silvereel[2/]<br>Bluerunner<br>Red Drum<br>Shark<br>Spanish Mackerel |

1/ One set was 40 tons

2/ Accounted for 69% of T. catch

# ADDUCTOR MUSCLE PARASITES, SULCASCARIS SULCATA, IN CALICO SCALLOPS FROM THE SOUTHEAST COAST OF THE UNITED STATES

N.J. Blake, B.J. Barber, G.E. Rodrick, and C.D. Burns
Department of Marine Science, University of South Florida
140 Seventh Avenue South, St. Petersburg, Florida 33701

## ABSTRACT

In late 1981 and 1982 the value of the calico scallop (Argopecten gibbus) industry, centered off the east coast of Florida, was endangered as a result of the occurrence and recognition of parasites, presumably Sulcascaris sulcata, in the adductor muscle. In order to determine the geographic distribution of the parasites along the southeast coast of the United States, scallops were collected in June 1982 from a total of 16 stations ranging from Savannah, Georgia to Fort Pierce, Florida.

Occurence of at least one parasite per muscle ranged from 28 to 68% with a mean of 45.5%. Both third and fourth stage larvae were present. A latitudinal pattern of parasite occurrence was not observed; neither were levels of parasite occurrence significantly correlated with scallop size.

## INTRODUCTION

In late 1981 and early 1982 the value of the calico scallop (Argopecten gibbus) industry, centered off the east coast of Florida, was endangered as a result of the occurrence and recognition of a larval nematode, presumably Sulcascaris sulcata, in the edible adductor muscle. The U.S. Food and Drug Administration ruled that not more than 20 out of 100 meats could contain a detectible parasite. One processor had to discard 13,000 lbs. of processed meats when they were found to contain the number of parasites which exceeded the allowable 20% level set by the regulatory agency.

The calico scallop industry has grown rapidly over the past few years and this rapid growth has undoubtedly led to some of the regulatory agency's initiative. The occurrence of these larval parasites in marine molluscs is a widespread phenomenon. Larval Sulcascaris have been reported in at least 12 species of marine bivalves and gastropods (5), many of which have commercial value. In spite of its occurrence in commercial species, there is no evidence that the parasite represents a

human health hazard.

The present study was undertaken to determine the range of occurrence levels in calico scallops during the summer along the southeast coast of the United States.

## MATERIALS AND METHODS

The locations of calico scallop beds fluctuate annually and to some extent seasonally depending upon where the larvae set. Historically, the scallop beds off the southeast coast of the United States have ranged from North Carolina to Stuart, Florida, with heaviest concentrations usually in the vicinity of Cape Canaveral (3).

In June 1982 during a cruise on R/V DELAWARE II, a total of 106 stations were sampled from near Savannah, Georgia to Fort Pierce, Florida. Sixteen of these stations (Fig. 1) yielded at least one bushel per station of scallops after a 1/2 hour tow with an 8 foot tumbler trawl. One hundred scallops were randomly selected from each sample and the maximum length (dorsal-ventral) of each scallop was measured to the nearest 0.1 mm. Fifty of these scallops were then dissected and individual adductor muscles were placed in plastic bags and frozen for later analysis.

Upon returning to shore, the muscles were thawed and the exterior of each muscle was examined for the presence of parasites under a table-mounted illuminated magnifier. Each muscle was then squashed between two glass plates and again examined under the magnifier. The exterior number of parasites in each muscle was subtracted from the total number of parasites in order to obtain the interior occurrence levels.

## RESULTS AND DISCUSSION

The various larval stages and hosts of Sulcascaris sulcata in Australian waters have been delineated (1, 2). The adult Sulcascaris sulcata in the stomachs of marine turtles produce eggs which are excreted and deposited on sediment. After two molts within the egg, a third larval stage emerges which infects a bivalve or gastropod. Three to four months later a fourth larval stage develops which is about 5 mm long and unless the mollusc is eaten by a turtle, continues to grow to almost 30 mm by the end of six months. The developmental stages of the parasite from the waters off the southeast coast of the United States are probably similar although the duration spent in each larval stage are probably somewhat different.

In the present study, relatively few third stage larvae were observed. Of the 530 parasites counted, only 34 were third stage, while the remainder were fourth stage. It's possible that the low number of third stage larvae was due to

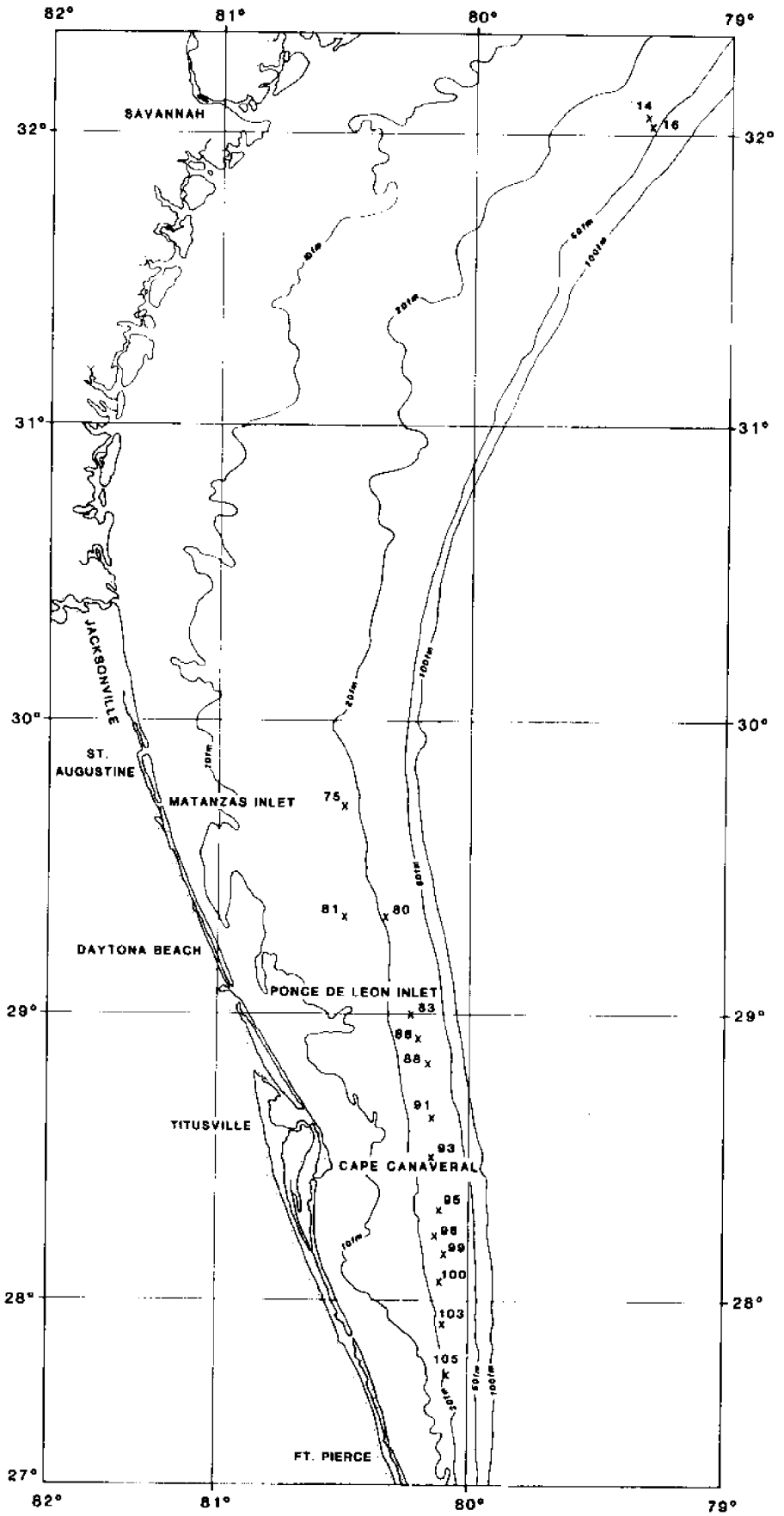



Figure 1. Calico scallop sampling locations.

the breeding cycle of the parasite. More third stage larvae might be encountered during the spring.

Table 1 shows the occurrence levels of Sulcascaris in the adductor muscles of scallops from the 16 stations. External percent occurrence ranged from 10% to 34% and internal percent occurrence ranged from 16% to 54%. Internal percent occurrence levels were almost always greater than the external levels. Combined percent occurrence levels ranged from 26% to 68% with a mean of 45.5%. These levels are similar to those reported for two collections of calico scallops made in 1970 and 1976 off Cape Canaveral (5). Thus, none of the 16 stations sampled during June 1982 could meet the FDA requirement of less than 20% occurrence.

The station numbers shown in Table 1 increase from north to south. During this June sampling a latitudinal trend in occurrence levels was not observed. For example stations 14 and 16 in the north showed combined occurrence levels of 16% and 48%, respectively, while stations 100 and 103 in the south showed combined levels of 24% and 42%, respectively.

Station 105 near Fort Pierce had the highest combined occurrence level of 68%. In surf clams (Spisula solidissima) from Virginia, the occurrence level of this parasite has been shown to be as high as 78% (4).

The percent occurrence levels were poorly correlated ($r = .41$) to mean shell length (Fig. 2). The mean shell length of scallops from the 16 stations ranged from 43.2 mm to 54.3 mm. Larger scallops, however, did not necessarily have the higher occurrence levels. The poor correlation is the result of the wide range in occurrence levels for any particular size of scallop. It's possible that a significant correlation with size could be found if the size range of the scallops collected were expanded and more points could be added.

## CONCLUSION

During the June 1982 sampling of 16 stations along the southeast coast of the United States, combined internal and external levels of third and fourth stage larvae of Sulcascaris sulcata in the adductor muscle of calico scallops, always exceeded the 20% occurrence level mandated by the Food and Drug Administration as the upper acceptible limit. Levels did not appear to be well correlated with scallop size and the lack of latitudinal trends suggests a broad distribution.

## ACKNOWLEDGEMENTS

This work was supported by the Florida Sea Grant, National Oceanographic Administration of Sea Grant, Department of Commerce under grant No. 125720073/257*V73. The authors also

Table 1. Occurrence levels of Sulcascaris sulcata larvae in the adductor muscle of calico scallops from the southeast coast of the United States.

| | STATION # | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 14 | 16 | 75 | 80 | 81 | 83 | 86 | 88 | 91 | 93 | 95 | 96 | 99 | 100 | 103 | 105 |
| EXT % OCCURRENCE | 16 | 34 | 20 | 26 | 20 | 12 | 26 | 34 | 10 | 28 | 24 | 14 | 16 | 14 | 28 | 34 |
| INT % OCCURRENCE | 16 | 44 | 32 | 40 | 36 | 20 | 32 | 28 | 24 | 42 | 36 | 18 | 22 | 26 | 44 | 54 |
| COMBINED % OCCURRENCE | 30 | 62 | 48 | 60 | 46 | 28 | 44 | 52 | 32 | 54 | 48 | 26 | 34 | 34 | 62 | 68 |
| TOTAL # PARASITES/ 50 SCALLOPS | 16 | 48 | 32 | 38 | 35 | 20 | 41 | 34 | 21 | 41 | 41 | 17 | 20 | 24 | 42 | 60 |
| MEAN SHELL LENGTH (mm) | 44.3 | 54.3 | 50.1 | 53.6 | 51.8 | 48.9 | 44.2 | 43.2 | 43.2 | 48.5 | 48.1 | 48.0 | 49.2 | 44.7 | 46.2 | 48.9 |

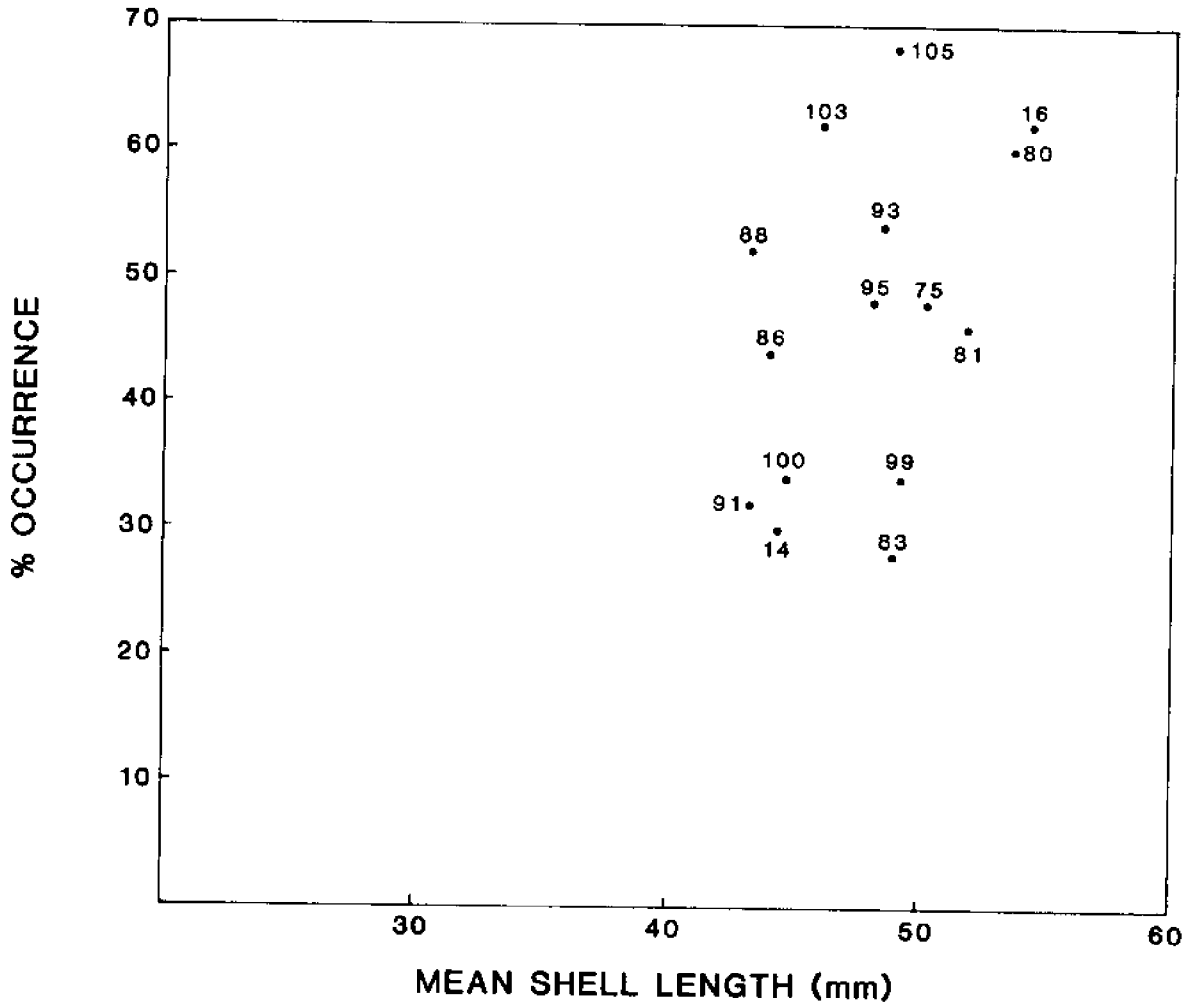



Figure 2. Combined internal and external occurrence levels of Sulcascaris sulcata in the adductor muscle of calico scallops in relation to shell size.

wish to thank the National Marine Fisheries Service and the Florida Department of Natural Resources for providing shiptime on R/V DELAWARE II.

## REFERENCES

1. BERRY, G. N. and L. R. G. CANNON. 1981. The life history of Sulcascaris sulcata (Nematoda: ascaridiodea), a parasite of marine molluscs and turtles. International Journal of Parasitology 11: 43-54.

2. CANNON, L. R. G. 1978. A larval ascaridoid nematode from Queensland scallops. International Journal of Parasitology 8: 75-80.

3. CUMMINS, R. 1971. Calico scallops of the southeastern United States, 1959-69. Special Scientific Report - Fish. No. 627. N.O.A.A., Seattle, 22 pp.

4. KERN, F. G., III. 1977. Distribution and prevalence of larval anisakid nematodes in surf clams from the Middle Atlantic Coast. Abstract, 52nd Meeting of the American Society of Parasitologists, August 14-19, Las Vegas, p. 53.

5. LICHTENFELS, J. R., T. R. SAWYER, and G. C. MILLER. 1980. New hosts for larval Sulcascaris sp. (Nematoda, anisakidae) and prevalence in the calico scallop (Argopecten gibbus). Trans. Amer. Micros. Soc. 99(4): 448-451.

# DETERMINATION OF THE THERMAL DEATH TIME OF *VIBRIO CHOLERAE* IN SHRIMP (*PENAEUS SETIFERUS*)

Arthur Hinton, Jr. and Robert M. Grodner
Department of Food Science
Louisiana Agricultural Experiment Station
Agricultural Center
Louisiana State University
Baton Rouge, Louisiana 70803

## INTRODUCTION

Since 1973 several cases of *Vibrio cholerae* 01 infections have been reported in the United States (6). Seafood was implicated as the source of the organism in some of these cases, and it has been responsible for cholera epidemics occurring in other countries (1,2,3).

Cooked crabs were implicated in 11 cases of the disease which occurred in Louisiana during 1978 (10). Authorities suspect that the organism either survived the cooking or that the cooked crabs were recontaminated when they were returned to their original container for storage. After this outbreak, hundreds of samples of fresh Louisiana crabs, oysters, and shrimp were examined. *V. cholerae* was isolated from shrimp caught in a canal near one of the sites where the implicated crabs were caught (2).

Although it is generally recognized that *V. cholerae* is a very fragile organism (9), relatively little information has been published on the thermal resistance of *V. cholerae* 01. The purpose of this study was to determine the thermal resistance of the organism in shrimp meat.

## MATERIALS AND METHODS

*Organism.* *Vibrio cholerae* 01 (Biotype El Tor, Serotype Inaba) Louisiana Strain #5875 was obtained from the Department of Microbiology at Louisiana State University. The organism was stored on peptone salt agar slants at room temperature and transferred to fresh media at 4 to 6 weeks intervals.

*Preparation and inoculation of shrimp homogenate.* Fresh shrimp were purchased from local seafood stores. Three parts peeled shrimp and 1 part distilled water were blended at high speed in a Waring blender until smooth.

*V. cholerae* grown in nutrient broth for 20 to 24 hr at 35°C

was added to the homogenate to yield a final concentration of approximately $10^6$ organisms/g. The inoculated homogenate was throughly mixed by blending at high speed for 1 min.

Preparation of thermal death time tubes. Pyrex glass tubing with an outer diameter of 13 mm was cut in to 15 cm lengths and heat sealed at one end with an oxygen-acetylene torch.

Four grams of the shrimp homogenate were placed into the tubes with a sterile animal force feeder. The tubes were heat sealed with the oxygen-acetylene torch in lengths of 10 cm.

Determination of the come-up time. The come-up time of the homogenate was determined at each temperatures. Three tubes were connected to copper-constantan thermocouples and immersed in a water bath heated at the required temperature. The time required for the temperature of the samples to rise from room temperature to the water bath temperature was recorded on a Leeds and Northrup Speedomax multipoint recorder. The time required for the slowest heating tube to reach the water bath temperature was used as the come-up time for all other experiments at that temperature. The timing of the heat treatments was begun at the end of the come-up period. The lethal effect of the heat during the come-up time was determined mathematically (15).

Endpoint determination. Tubes containing the shrimp homogenate were immersed into a water bath heated from 48.9C (120F) to 82.2C (180F) at 4.6C (10F) intervals. The endpoint was bracketed by heating inoculated samples at 5 or 10 min intervals. One sample was removed at each interval and cooled in an ice water bath for 30 sec. The samples were tested for the presence of V. cholerae by an alkaline peptone water (APW) enrichment (16) and isolation of thiolsulfate cirtrate bile salts agar (TCBS) Difco). Typical V. cholerae colonies were confirmed biochemically and serologically. Cultures which produced the proper reactions of Klieger's iron agar (BBL)and lysine iron agar (Difco) and were agglutinated by Bacto Vibrio cholerae antiserum, Polyvalent and Inaba (Difco), were confirmed as V. cholerae.

Sets of 15 thermal death time tubes were then heated at 1 min intervals above and below the time which produced negative results in the preliminary one tube tests. The time period after which no V. cholerae was recovered from any of the 15 tubes of heated homogenate was defined as the endpoint.

The control sample consisted of inoculated homogenate which underwent the same treatment as the other experimental sample, except no heat treatment was applied.

Inoculated pack studies. Fresh peeled shrimp were injected with a 20 to 24 hr V. cholerae culture grown at 35C in nutrient broth. A syringe was used to inject $10^6$ V. cholerae into the shrimp.

Fifty gram samples of the injected shrimp were cooked using one of two methods. One group of shrimp was cooked in a liter of boiling water for 10 min, and the other group was cooked in steam (100C, 212F) for 10 min.

After cooking 25g of the shrimp were blended in 225 ml of APW and incubated at 35C for 8 hr. Isolation and identification of V. cholerae were repeated as described above. A control sample consisting of inoculated shrimp was treated in the same manner as above, exception that the shrimp were not cooked.

## RESULTS AND DISCUSSION

Recovery of V. cholerae from the homogenate. Death of the organism was defined as the inability of the bacteria to form colonies on TCBS after enrichment in APW; therefore, it was important that a large percentage of the live organisms be recovered from the homogenate. An average of 70% of the organisms was recovered from the unheated homogenate with the Most Probable Number (MPN) technique (Table 1).

Endpoints. V. cholerae was not recovered from any of 15 tubes containing the inoculated homogenate which were heated for 69 min or more after reaching 48.9C (120F) Table 2). At this temperature, 4.17 min were required for the temperature of the homogenate in the tubes to rise from room temperature to the temperature of the water bath. This was equivalent to heating the homogenate at that temperature for 2.16 min (Table 2) which produced a total heating time of 71.16 min (Table 3).

V. cholerae was not recovered from the homogenate heated for 1 min after reaching the temperatures from 54.4C to 82.2C. The corrected endpoints at these temperatures ranged from 3.34 to 2.20 min (Table 3). The minimal heating time used in these studies was 1 min. However, no organisms could be recovered from any of the thermal death time tubes after 1 min of heating at 54.4C or above. This meant that the true endpoints for these temperatures were less than 1 min. Therefore, the true corrected endpoints and the calculated D values were less than their calculated values.

D values. D values for each temperature were calculated using the corrected endpoints. The values ranged from 9.17 min at 48.9C to less than 0.28 min at 82.2C (Table 3).

Heat treatments which produce a 4 to 7 log cycle reduction of the bacterial population are generally considered adequate for pasteurization (13). The amount of time required to produce a 7 log reduction of the V. cholerae population in the shrimp homogenate was calculated by multiplying the time for 1 log cycle by 7 (Table 3).

V. cholerae is very heat sensitive. It has been reported that the organism is killed after 10 min at 55C (14). That study was done with the classical biotype, not with the more resistant El Tor

Table 1

EFFICIENCY OF RECOVERY OF VIBRIO CHOLERAE FROM UNHEATED SHRIMP HOMOGENATE

| | Trial 1 | Trial 2 | Trail 3 |
|---|---|---|---|
| ORGANISMS PLACED IN SHRIMP HOMOGENATE | 1.53 X $10^6$/g | 2.80 x $10^6$/g | 9.78 X $10^5$/g |
| ORGANISMS RECOVERED FROM SHRIMP HOMOGENATE | 4.6 X $10^5$/g | 2.4 x $10^6$/g | 9.3 X $10^5$/g |
| PERCENTAGE OF ORGANISMS RECOVERED | 30% | 85.7% | 95% |

Table 2

ENDPOINTS OF V. CHOLERAE IN SHRIMP MEAT HOMOGENATES AND HOMOGENATE COME UP TIME

| TEMPERATURE (°C) | ENDPOINT[a] (MIN) | COME UP TIME (MIN) | LETHALITY OF COME UP TIME (MIN) |
|---|---|---|---|
| 48.9 | 69 | 4.17 | 2.16 |
| 54.4[b] | 1 | 4.34 | 2.34 |
| 60.0[b] | 1 | 3.02 | 2.02 |
| 65.5[b] | 1 | 2.51 | 1.51 |
| 71.1[b] | 1 | 2.39 | 1.39 |
| 76.7[b] | 1 | 2.34 | 1.34 |
| 82.2[b] | 1 | 2.20 | 1.20 |

[a]ENDPOINTS BASED ON RECOVERY OF NO V. CHOLERAE FROM 15 TUBES CONTAINING A TOTAL OF $6.0 \times 10^7$ ORGANISMS.

[b]TRUE D VALUES OBTAINED AT THESE TEMPERATURES ARE LESS THAN THOSE GIVEN DUE TO THE MINIMAL TEST PERIOD OF 1 MIN.

Table 3

CORRECTED ENDPOINTS AND D VALUES
FOR V. CHOLERAE IN SHRIMP HOMOGENATE

| TEMPERATURE ($^{O}$C) | CORRECTED ENDPOINTS (MIN) | D VALUES (MIN) | 7D VALUES (MIN) |
|---|---|---|---|
| 48.9 | 71.16 | 9.17 | 64.02 |
| 54.4[a] | 3.34 | 0.43 | 3.00 |
| 60.0[a] | 3.02 | 0.39 | 2.73 |
| 65.5[a] | 2.51 | 0.32 | 2.24 |
| 71.1[a] | 2.39 | 0.31 | 2.17 |
| 76.7[a] | 2.34 | 0.30 | 2.10 |
| 82.2[a] | 2.20 | 0.28 | 1.96 |

[a]TRUE D VALUES OBTAINED AT THESE TEMPERATURES ARE LESS THAN THOSE GIVE DUE TO THE MINIMAL TEST PERIOD OF 1 MIN

biotype however (12).

Many factors influence the thermal resistance of bacteria (5). At 48.9C V. cholerae #5875 has a D value of 1.70 min in peptone water. 4.72 min in oyster meat homogenate (8), and 8.15 min in crab meat homogenate (13). The D value of the organism was greater in the meat homogenates than in the peptone water. The presence of colloidal substances such as proteins and fats may enhance the resistance of the organism in the meat homogenates (4).

V. cholerae #5875 is more heat resistant in shrimp meat homogenate than in the oyster and crab meat homogenates. This may be related to the pH of the meat homogenates. Generally, bacteria are most heat resistant in a substrate that is at or near neutrality (4). Acid and alkaline conditions decrease the heat resistance of bacteria, but a change in pH toward acidity causes a greater reduction in the heat resistance than a corresponding increase in alkalinity. The pH of the shrimp homogenate was between 7.2 and 7.9. The pH of fresh crab meat is between 7.2 and 7.4, and the pH of the oyster homogenate was 6.0 (8). The viability of V. cholerae is reportedly less in an environment with a pH less than 6.2 (11).

Inoculated packs. The shrimp which were steamed weighed between 2.1 and 21.7g (Table 4). The weight of the shrimp determined the rate of heat penetration to the cold point of the shrimp. The temperature of shrimp weighting 7g or less is 100C after cooking for 2 min in 100C steam (7), but the temperature of shrimp weighing 13.6g is 96.67C after the same time. No V. cholerae was recovered from any of the shrimp cooked for 10 min with 100C steam (Table 4).

The results obtained when inoculated shrimp weighing 2.8 to 23.2g were cooked in boiling water are shown in Table 5. It requires approximately 4 min for the internal temperature of a 10g shrimp to reach 100C (7). More time would be required for heavier shrimp. The results of cooking the shrimp in boiling water for 10 min were the same as those obtained by cooking the shrimp with steam. Both methods were effective for destroying $10^6$ V. cholerae/shrimp.

## REFERENCES


1. BAINEW, W.B., A. ZAMPEIRI, M. MAZZOTI, G. ANGIONI, M.S. GOILIA, E. IZZO, F. GANGAROSO, and F. POCHIARI. 1974. Epidemiology of Cholera in Italy in 1973. Lancet ii. 1370-1374.

2. BLAKE, P.A., D.T. ALLEGRA, J.D. SNYDER, T.J. BARRET, L. MCFARLAND, C.T. CARAWAY, J.C. FEELEY, J.P. CRAIG, G.V. LEE, N.D. PAHR, and R.A. FELDMAN. 1980. Cholera- a possible endemic focus in the United States. New England J. Med. 302:305-309.

Table 4

SURVIVAL OF V. CHOLERAE IN SHRIMP
STEAMED AT 100C (212F) FOR 10 MINUTES

| SAMPLE | WEIGHT OF SHRIMP (GRAMS) | RESULTS |
|---|---|---|
| 1 | 2.10-3.99 | -[a] |
| 2 | 3.30-13.66 | -[a] |
| 3 | 4.46-21.74 | - |
| UNCOOKED | NOT WEIGHED | +[b] |

[a] NO V. CHOLERAE RECOVERED FROM SAMPLE

[b] V. CHOLERAE RECOVERED FROM SAMPLE

Table 5

SURVIVAL OF V. CHOLERAE IN SHRIMP
COOKED IN WATER AT 100C (212F) FOR 10 MINUTES

| SAMPLE | WEIGHT OF SHRIMP (GRAMS) | RESULTS |
|---|---|---|
| 1 | 2.80-3.80 | -[a] |
| 2 | 4.30-11.30 | -[a] |
| 3 | 4.30-23.20 | - |
| UNCOOKED | NOT WEIGHED | +[b] |

[a] NO V. CHOLERAE RECOVERED FROM SAMPLE

[b] V. CHOLERAE RECOVERED FROM SAMPLE

3. BLAKE, P.A., J. ROSENBERG, J. COSTA, P.S. FEREIRA, C.L. GUEMARAES, and E.J. GANGAROSA. 1977. Cholera in Portugal, 1974. Modes of transmission. Am. J. Epidemiol. 105:337-343.

4. FRAZIER, W.C. and D.C. WESTHOFF. 1978. Food Microbiology. McGraw-Hill Book Co. New York.

5. HANSEN, N.N. and H. RIEMAN. 1963. Factors affecting the heat resistance of non-sporing organisms. J. App. Bacteriol. 26:314-333.

6. HELTON, B., W.E. BIRCH, J. HOLDEN, W.E. PERCY, H.B. BRADFORD, R. CONLEY, L.M. MCFARLAND, J.R. ROMERO, C.T. CARAWAY, C. DAVIS, A. OCHOA, D.C. BLAKEY, V. BATEMAN, P.M. FORTNEY, L. HAGGARD, A. ARCALA, R. CROSSMAN, D. MARTIN, D. MASSESANG, J. PERDUE, and C.R. WEBB, JR. 1981. Cholera on a Gulf coast Oil Rig. CDC Morbid, Mortal. Weekly Rep. 30-589-590.

7. HIMELBLOOM, B.H. 1978. Heat penetration, shell color changes, and meat yields of crabs, crayfish, and shrimp under various cooking conditions. M.S. Thesis. Louisiana State University. Baton Rouge, LA.

8. HINTON, A., JR. and R.M. GRODNER. 1982. Determination of the thermal death time of Vibrio cholerae of oysters. (Crassostrea virginica). Presented at the Seventh Annual Tropical and Subtropical Fisheries Technological Conference of the Americas. New Orleans, LA.

9. HIRSCHHORN, N. and W.B. GREENOUGH III. 1971. Cholera. Sci. American 225:15-21.

10. LOUISIANA MONTHLY MORBIDITY REPORT. Sept. 1978. Dept. of Health and Human Resources, New Orleans, LA.

11. OUCHTERLONY, O. 1958. A selective medium for isolating cholera vibrios. Acta. Path. Microbiol. Scand. 43:274-284.

12. SAKAZAKI, R. 1979. Vibrio infections. p. 174-209. In: H. Rieman and F.L. Bryan (eds.), Foodborne infections and intoxications. Academic Press. London.

13. SCHULTZ, L.M., R.M. GRODNER, S.L. BIEDE, and J.E. RUTLEDGE. 1980. Determination of ther thermal death time of Vibrio cholerae in crab meat homogenate. Presented at the 77th Meeting fo the Southern Association of Agric. Scientist. Hot Springs, Arkansas.

14. SMITH, D.T. and N.F. CONANT. 1957. Vibrio comma and asiatic cholera. In: Zinsser Microbiology. Appleton-Century-Crofts. New York.

15. STUMBO, C.R. 1965. Thermobacteriology in Food Processing. Academic Press. New York.

16. U.S.F.D.A., BUREAU OF FOODS, DIVISION OF MICROBIOLOGY. Bacteriological Analytical Manual. Isolation and identification of Vibrio cholerae. Association of Analytical Chemist. Washington, D.C.

# EVALUATION OF THE ELEVATED TEMPERATURE INCUBATION PROCEDURE FOR THE RECOVERY OF SALMONELLAE FROM OYSTERS

Mary L. Miller and John A. Koburger
Food Science and Human Nutrition
University of Florida
Gainesville, Florida 32611

## INTRODUCTION

The isolation of low levels of salmonellae from food and water samples is often complicated by the presence of competing microorganisms. Several laboratories have observed that the use of elevated temperatures, i.e., 41-43C, lessened the competition and allowed for improved isolation of salmonellae (4, 5, 17). Increased recovery of salmonellae by incubation of selective enrichment broths at 43C rather than 35-37C was first reported by Harvey and Thompson (12). Georgala and Boothroyd (9) studied direct enrichment in Selenite F broth at 43C and 37C on meat and meat products and found the best recovery of salmonellae was at 43C. Hobbs (13) found incubation at 35-37C to be better for the isolation of salmonellae than at higher temperatures, but various laboratories have confirmed the advantages of using elevated temperatures (14, 16, 17). However, it should be noted that certain serotypes such as S. typhi and S. pullorum are known not to grow at 43C, and temperatures above 43C are inhibitory to salmonellae growth and recovery (11).

Selenite and Tetrathionate broths are the selective enrichment media of choice for salmonellae with preenrichment in Lactose broth usually recommended (1, 7). Silliker and Gabis (8) found that preenrichment followed by selective enrichment resulted in increased recovery of salmonellae from raw meats. Other researchers (3, 6, 10) have also observed that preenrichment allows for the best recovery of salmonellae as it enables any injured bacteria time to repair before being subjected to a selective media. However, it has been recommended that raw or highly contaminated products not be preenriched (7) because of the opportunity for overgrowth of the salmonellae by competing microorganisms.

The incubation period for selective enrichment broths varies from lab to lab with the most common periods being 24 and 48 hrs. Kafel and Bryan (14) found no difference in recovery between 24 and 48 hr incubation at 37C, but at 43C, the 48 hr incubation appeared to be better. Other researchers have found that longer incubation with multiple subcultures increases the recovery of salmonellae (15).

A combination of selective plating media has been advised (11) to ensure isolation of all groups of salmonellae. Andrews et al. (2)

studied 5 selective plating agars in various combinations on a number of food types. They indicated that Xylose Lysine Desoxycholate, Bismuth Sulfite and Brilliant Green agars were effective with almost 100% recovery of salmonellae when used in combination.

The purpose of this study was to investigate the use of elevated temperatures in conjunction with preenrichment, direct selective enrichment, selective plating media and incubation time on the isolation of salmonellae from the oyster, Crassostrea virginica.

## MATERIALS AND METHODS

### Oyster Samples

Five oyster samples were harvested in the area of Suwannee, Florida, and two samples were obtained at retail for a total of seven samples. The oysters were brushed clean in running tap water and shucked into a sterile blender jar. A composite sample of 40 or more oysters was obtained by blending the oysters for 2 min in a Waring Blender. Subsamples of 25 gr each were then diluted 1:10 in Lactose broth, Selenite Cystine, (SC), and Tetrathionate broth, (TT). Each of the 7 composites were analyzed in triplicate.

### Preenriched Samples

Three aliquots from each sample were incubated in lactose broth at 35C for 24±2 hr, then 1 ml from each sample was introduced into triplicate 10 ml tubes of SC and TT. One set of tubes was incubated at 35, one at 41 and the final set at 43C, each for 24±2 hr.

### Selectively Enriched Samples

Three aliquots from each sample were diluted 1:10 directly into the selective enrichment broths, SC and TT, without first being preenriched. The samples were incubated at 35, 41 and 43C for 24±2 and 48±2 hr.

### Plating Media

After incubation, each of the selective broths was streaked onto Xylose Lysine Desoxycholate (XLD), Bismuth Sulfite (BS), and Brilliant Green (BG) agars. All plates were incubated at 35C for 24 hr. Observations were also made regarding the amount of non-Salmonella colonies developing on the plates.

### Identification

Two colonies from each plate that appeared typical of salmonellae were transferred to Triple Sugar Iron agar (TSI) and Lysine Iron agar (LIA). Those isolates which were typical of salmonellae were purified

and then identified by conventional biochemical (7) and slide agglutination tests using Salmonella O antiserum, Polyvalent A-I and Vi (Difco, BBL).

## RESULTS AND DISCUSSION

Preliminary studies in our laboratory had established that low levels of salmonellae were present in oysters harvested in certain approved locations in the area of Suwannee, FL. It was, therefore, decided to collect samples from this source rather than to use laboratory contaminated samples. There are a number of inherent problems associated with the use of laboratory cultures in recovery studies, such as adjustment of inoculum levels and the failure of laboratory cultures to respond to environmental stress as do the wild-types. In addition two retail samples of oysters were obtained and analyzed.

The number of samples positive for salmonellae generally increased with the temperature of incubation of the selective enrichment media as shown in Table 1. The number of samples positive for salmonellae was the same at 41 and 43C, but the number of isolates obtained at 43C was greater. Direct selective enrichment yielded more positive samples (30 vs 14) and more isolates than the lactose preenrichment with SC yielding the greatest number of positive samples. This was somewhat surprising in that preenrichment is now recommended for most samples (1) in order that debilitated cells have a opportunity to undergo repair.

Table 1. Number of Salmonella positive aliquots, isolates recovered, and the effect of selected enrichment as influenced by incubation temperature

| | Lactose Preenriched | | | Direct Selective Enrichment | | |
|---|---|---|---|---|---|---|
| | 35C | 41C | 43C | 35C | 41C | 43C |
| Positive Aliquots | 3/21[a] | 4/21 | 7/21 | 8/21 | 11/21 | 11/21 |
| Positive Isolates | 6 | 11 | 19 | 16 | 44 | 70 |
| Positive Samples From: | | | | | | |
| Selenite Crystine | 2/21 | 2/21 | 0/21 | 4/21 | 9/21 | 8/21 |
| Tetrathionate Broth | 3/21 | 4/21 | 7/21 | 4/21 | 2/21 | 3/21 |

a: Number aliquots positive for salmonellae/total aliquots tested

Direct selective enrichment in SC resulted in better recovery of salmonellae than in TT at the temperatures of 41 and 43C. The lactose preenriched samples showed the opposite effect with almost all the isolates being recovered from TT. This effect was again enhanced by elevated temperatures with 100% of the 43C isolates coming from TT following lactose preenrichment. At 35C direct enrichment in SC and TT were about equally effective in recovery of salmonellae. These data may be the result of a selective process on serotypes due to temperature as well as media.

No single plating media, i.e., XLD, BS or BG, proved superior to another in recovering salmonellae. However, XLD did appear to be more effective with lactose preenriched samples at elevated temperatures, but this may be due to ease of recognition rather than improved recovery. The use of a combination of plating media increases the chance of recovery of salmonellae and is widely recommended (1, 2, 7).

The incubation period was a factor only in the direct selective enrichment samples (no data shown). At elevated temperatures 48 hr incubation was slightly favored; whereas, 24 hr incubation was slightly favored at 35C. Extended incubation, however, did not increase the number of samples that were positive.

In general, salmonellae were isolated from oysters more often from direct selective enrichment in SC at elevated temperatures. This effect was observed with samples containing naturally occurring Salmonella and was unexpected in light of the reported superiority of TT following preenrichment for the recovery of salmonellae (3, 10). The main advantage of using elevated temperature incubation appears to be in the reduction of competing microorganisms during growth, thereby, allowing for the more efficient isolation of the salmonellae. This observation has been reported previously (11).

These results demonstrate the improved isolation of naturally occurring populations of salmonellae in oysters at elevated temperatures. Whether there is a recognizable selection of serotypes due to the use of these various media at elevated temperatures needs to be investigated further.

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REFERENCES

1. American Public Health Association. 1976. Compendium of methods for the microbiological examination of foods. Ed., M. L. Speck. American Public Health Association, Washington, DC.

2. Andrews, W. H., C. R. Wilson, P. L. Poelma, and A. Romero. 1979. Microbiological methods: Relative productivity of five selective plating agars for the recovery of Salmonella from selected food types. J. Assoc. Off. Anal. Chem. 62:320-326.

3. D'Aoust, J. 1981. Update on preenrichment and selective enrichment conditions for detection of Salmonella in foods. J. Food Protect. 44:369-374.

4. Edel, W. and E. H. Kampelmacher. 1968. Comparative studies on Salmonella isolation in eight European laboratories. Bull. W.H.O. 39:487-491.

5. Edel, W. and E. H. Kampelmacher. 1969. Salmonella isolation in nine European laboratories using a standardized technique. Bull. W.H.O. 41:297-306.

6. Edel, W. and E. H. Kampelmacher. 1973. Comparative studies on isolation methods of 'sub-lethally' injured salmonellae in nine European laboratories. Bull. W.H.O. 48:167-174.

7. Food and Drug Administration. 1978. Bacteriological Analytical Manual, 5 ed. Chapter 6, pp. 1-29, AOAC, Washington, DC.

8. Gabis, D. A. and J. H. Silliker, 1974. ICMSF methods studies II. Comparison of analystical schemes for detection of Salmonella in high-moisture foods. Can. J. Microbiol. 20:663-669.

9. Georgala, D. L. and M. A. Boothroyd. 1965. A system for detecting salmonellae in meat and meat products. J. Appl. Bacteriol. 28:206-212.

10. Harvey, R. W. S. and T. H. Price. 1977. Observations on preenrichment for isolating salmonellaes from sewage polluted natural water using Muller-Kauffman tetrathionate broth prepared with fresh and desiccated ox bile. J. Appl. Bacteriol. 43:145-148.

11. Harvey, R. W. S. and T. H. Price. 1979. A review: Principles of Salmonella isolation. J. Appl. Bacteriol. 46:27-56.

12. Harvey, R. W. S. and S. Thompson. 1953. Optimum temperature of incubation for isolation of salmonellae. Mon. Bull. Ministr. Health Public Lab Serv. 12:149-150.

13. Hobbs, B. C. 1962. Chemical and biological hazards in foods. Edited by J. C. Ayres, A. A. Kraft, H. E. Snyder, and H. W. Walker. Iowa State University Press, Ames.

14. Kafel, S. and F. L. Bryan. 1977. Effects of enrichment media and incubation conditions on isolating salmonellae from ground-meat filtrate. Appl. Environ. Microbiol. 34:285-291.

15. Morris, G. K. and C. G. Dunn. 1970. Influence of incubation temperature and sodium heptadecyl sulfate (Tergitol No. 7) on the isolation of salmonellae from pork sausage. Appl. Microbiol. 20:192-195.

16. Silliker, J. H. and P. A. Gabis, 1974. ICMSF methods studies V. The influence of selective enrichment media and incubation temperatures on the detection of salmonellae in raw frozen meats. Can. J. Microbiol. 20:813-816.

17. Spino, D. F. 1966. Elevated-temperature technique for the isolation of salmonella from streams. Appl. Microbiol. 14:591-596.

# INCIDENCE OF SALMONELLAE IN CLAMS, OYSTERS, CRABS AND MULLET HARVESTED FROM TWO LOCATIONS IN FLORIDA

M. B. Fraiser and J. A. Koburger
Food Science and Human Nutrition
University of Florida, Gainesville, FL 32611

## INTRODUCTION

The reported incidence of salmonellosis in humans has been increasing steadily in recent years (5) as the general public and public health officials become more aware of the disease and recognition of the organism is made easier by improved isolation methods. Contamination of seafoods with salmonellae has been a particular problem within the industry, as the potential for sewage and other discharges entering seawater becomes more prevalent. To complicate the control of Salmonella as well as the monitoring of harvest waters, the usefulness of coliform organisms as indicators of pollution has been recently questioned (1, 2, 8, 11, 12). In addition, mollusks are known to concentrate organisms within their bodies, thus rendering analysis of the accompanying water of questionable value (18).

Seafoods were responsible for 8.7% of the foodborne disease outbreaks in the United States in 1979 (3). It is unknown in many instances whether these outbreaks were due to processing failures and poor handling procedures or whether the outbreaks were due to pathogens that are members of the normal flora of the seafood, as was recently found with Vibrio cholerae (6, 10). With the dockside value of seafoods landed in Florida at approximately 173 million dollars in 1981 (15), it would be advantageous to study the incidence of Salmonella in raw seafood and be aware of their presence before processing failure or mishandling could occur.

The purpose of this research was to study the incidence of Salmonella in four seafoods commercially harvested in Florida: oysters (Crassostrea virginica), clams (Mercenaria mercencaria), striped mullet (Mugil cephalus), and blue crabs (Callinectes sapidus). The seafoods were harvested from two areas approved for shellfish harvesting in Florida; the west coast location was at the mouth of the Suwannee River and the east coast location was in the Intracoastal Waterway at Crescent Beach.

## MATERIALS AND METHODS

Crabs were harvested by trapping, oysters by tonging, and clams by digging. The mullet were fresh commercial samples purchased in the vicinity of the sampling locations (Suwannee, FL and St. Augustine, FL) and were iced prior to transportation. Eighteen of the thirty east

coast crab samples were purchased live in St. Augustine and were not iced during transportation. All samples were transported to the laboratory in Gainesville in sanitized insulated coolers and analyses were begun within four hours of harvest. With the exception of the mullet, no attempt was made to cool the samples because of the short time span involved between collection and analysis. Thirty samples of each species from each location were analyzed individually for the presence of salmonellae. Aerobic plate counts, total coliform and fecal coliform analyses (19), were conducted using a composite sample for oysters and east coast clams, while crabs, mullet and west coast clams were not composited due to their larger size. Duplicate samples were anlyzed for aerobic plate counts, total coliforms, and fecal coliforms.

The method for isolation of salmonellae generally followed the Bacteriological Analytical Manual (19), which is briefly described as follows. Samples were blended for two minutes at 8000 rpm in a 1:10 dilution using lactose broth. Crabs weighing over 100 grams were mixed with lactose broth to a final volume of 900 grams. Mullet samples were not blended, but placed whole into individual plastic bags containing lactose broth in a 1:10 proportion. The bags were heat-sealed, the samples shaken, and the bags incubated. This method was used because of the larger size of the mullet and the desire to analyze the entire surface of the fish. All samples were incubated at 35 C for 24 ± 2 hours, followed by selective enrichment in both Selenite Cystine and Tetrathionate broths. Selective plating was performed using Bismuth Sulfite, Brilliant Green, and XLD agars, which was followed by transferring typical colonies to Triple Sugar Iron agar (TSI) and Lysine Iron agar (LIA) slants. After purification, biochemical tests (19) were performed as well as serological testing using polyvalent antisera. Specific serological identification was performed by the laboratories of the Florida Department of Health and Rehabilitative Services.

A quantitative storage study of salmonellae in oysters from the west coast location was performed using five 10-g samples and an appropriate MPN table (16). Composite samples of 100 grams were prepared and the analysis was performed in duplicate. Oysters were stored at 5C for up to 10 days.

Aerobic plate counts were incubated at 25 C for five days using Standard Plate Count agar. Analyses for coliform and fecal coliform organisms were performed using the Most Probable Number (MPN) technique as outlined in the Bacteriological Analytical Manual (19).

## RESULTS AND DISCUSSION

Although the total number of samples containing salmonellae (Table 1) seems surprisingly high at 33 out of 240 (13.4%), there are a number of factors which may have contributed to this. Higher rates of isolation

could have been the result of not using iced transportation, low numbers of competing microorganisms, the use of a preenrichment medium, and the analysis of individual animals rather than composite samples. Refrigeration is known to injure many microorganisms and because of this, no attempt was made to cool the samples with the exception of the mullet which were iced prior to purchase. The short period between harvest and analysis minimized any opportunity for extensive growth of competing microorganisms or major changes in the microbial population. Aerobic plate counts were low, indicating that competing microorganisms were not present in high enough numbers as to markedly inhibit the growth of salmonellae. A preenrichment procedure was also used, which is normally recommended for processed foods or foods with low levels of contamination (19). This use of a preenrichment medium allowed any injured organisms to overcome physiological stress. Because low numbers of salmonellae were anticipated, individual animals were analyzed so as to select a larger number of smaller sized samples, thereby increasing the probability of recovering any salmonellae present. If composite samples had been used, the probability of recovering all salmonellae present might have been reduced.

Table 1. Aerobic plate count, total coliform, fecal coliform and salmonellae recovered from four seafoods

| | | Coliforms | | | |
|---|---|---|---|---|---|
| | APC[1] | Total[1] | Fecal[1] | Salmonellae | |
| | | | | Samples pos./ | |
| Seafood | CFU/g | MPN/g | MPN/g | Samples tested | % |
| West Coast Oysters | 1100 | 21 | 2 | 3/30 | 10.0 |
| East Coast Oysters | 800 | 0 | 2 | 2/30 | 6.7 |
| West Coast Clams | 260 | 5 | 0 | 13/30 | 43.3 |
| East Coast Clams | $1100_5$ | 2 | 2 | 4/30 | 13.3 |
| West Coast Mullet | $4.4 \times 10^6$ | 78 | 0.2 | 0/30 | 0.0 |
| East Coast Mullet | $5.1 \times 10^5$ | 306 | 27 | 0/30 | 0.0 |
| West Coast Crab | $7.5 \times 10^5$ | 4700 | 4 | 11/30 | 36.7 |
| East Coast Crab | | | | | |
| Trapped | $7.9 \times 10^4$ | 0 | 0 | 3/12 | 25.0 |
| Purchased | $2.6 \times 10^8$ | 56 | 56 | 6/18 | 33.3 |

1: averages of duplicate determinations

The results indicate that salmonellae are widely distributed in the seafoods investigated. This is especially apparent as one considers the

two harvest locations. On the west coast, the area at the mouth of the Suwannee River is not highly populated, with only a limited number of riverfront vacation homes. The Intracoastal Waterway on the East Coast, however, is more populated as well as having sewage treatment plants which do not always function properly. It would seem, therefore, that if salmonellae were present because of fecal contamination, the east coast location would have a higher incidence of salmonellae. The opposite was found, with the highest incidence being in the west coast clams. Total coliform counts were also higher at the west coast location, yet the fecal coliform counts were higher at the east coast location (Table 1).

Table 2 lists the serotypes of Salmonella isolated in this study. As many as six different Salmonella species were isolated from the same sample lot indicating a rather wide variety of salmonellae in the environment. Only S. agona is listed among the ten most commonly reported species of salmonellae as being responsible for foodborne outbreaks in this country (4).

Table 2. Identification of Salmonella isolates from three seafoods

| | Number Samples Containing Serotype | | | | | |
|---|---|---|---|---|---|---|
| | Clams | | Oysters | | Blue Crabs | |
| Serotyes | West | East | West[a] | East | West | East |
| S. agona | | | | | 5 | 1 |
| S. allandale | | | | | | 1 |
| S. anatum | 1 | 1 | | 1 | | |
| S. bareilly | 2 | | | | | |
| S. braenderup | 2 | 1 | | | | |
| S. inverness | 1 | | 5 | | | 1 |
| S. java | 3 | | | 1 | | |
| S. muenchen | 8 | 1 | 3 | | | |
| S. redlands | | | | | | 2 |
| S. tallahassee | | 1 | | | 7 | 1 |
| S. thompson | | | 1 | | | |

[a]Isolates include those recovered during MPN study.

The mullet was the only species investigated that was negative for salmonellae, which may have been due to a number of factors. Mullet was the only free-swimming species studied, with the other species all being

"bottom dwellers." Stream bottom sediment has been found to have a higher recovery rate of salmonellae than surface waters (9), which would indicate that "bottom dwellers" would be more likely to become contaminated with salmonellae. Secondly, mollusks are filter feeders, which results in the accumulation of microorganisms from their environment (18), while fin fish are not filter feeders. Another possibility for the absence of salmonellae in mullet is the temperature of storage and incubation. The length of time the mullet were on ice prior to purchase is unknown. This icing may have been debilitating to any salmonellae on the fish. In addition, the total volume of fish and broth during the preenrichment of the mullet analysis was so great that several hours were required for all thirty samples to reach the incubation temperature of 35 C. Lastly, the method for analyzing the mullet differed in that the gastrointestinal tract was not exposed to the preenrichment medium. The plastic bag method was adopted to provide analysis of the total surface area of the fish. This approach to sampling was based on research involving sampling methods for detection of salmonellae in raw chicken carcasses (7). D'Aoust et al. found the whole carcass rinse method to be superior to thaw water analysis or the skin method for the detection of salmonellae.

Quantitation of the number of salmonellae per 100 g of oysters using the most probable number technique indicated very low numbers of salmonellae were present (Table 3). The highest number obtained was 2.2 salmonellae per 100 g (16). Because of the small size of the oysters (between 3 and 15 grams each), it would be possible for approximately one organism to be present in about every five oysters. With an infective dose being dependent upon the consumer, time factors, food preparation, and the nature of other foods consumed, it is highly unlikely that this concentration of salmonellae would cause gastroenteritis in humans.

Table 3. Most probable number of salmonellae present in west coast oysters

| Days of Storage | Sample | MPN per 100 g |
|---|---|---|
| 0 | A | 0.0 |
| | B | 2.2 |
| 5 | A | 2.2 |
| | B | 2.2 |
| 10 | A | 0.0 |
| | B | 2.2 |

It is also of interest to consider the survivability of salmonellae under commercial handling and storage conditions. The short storage study performed in conjunction with the MPN study showed that the salmonellae were able to survive at 5 C for 10 days. This, along with other studies (13, 14, 18), indicates that salmonellae can survive normal handling and storage procedures practiced in the seafood industry, although data regarding growth under these conditions are not available.

A relationship between the incidence of coliforms or fecal coliforms and salmonellae was not apparent ($r^2$ values at .15 and .02 respectively) as has been reported previously (1, 2, 17). With the exception of the mullet samples, total coliform counts seemed to parallel salmonellae incidence more closely than fecal coliform counts, but no obvious trend was apparent. This indicates the inadequacy of using indicator organisms as a measure of the safety of various species obtained from an estuarine environment. However, until a larger number of samples are analyzed, these data should be interpreted cautiously.

Although salmonellae were found in these seafood products, there is no indication that they present a public health problem. As the red meat and poultry industry became more cognizant of the level of salmonellae in these foods and was able to cope with this problem, a similar accommodation might be necessary with seafoods once a clearer knowledge of the source, levels and their fate in these foods is better understood.

## ACKNOWLEDGEMENTS

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## REFERENCES

1. Andrews, W. H., C. D. Diggs, J. J. Miescier, C. R. Wilson, W. N. Adams, S. A. Furfari, and J. F. Musselman. 1976. Validity of members of the total coliform groups for indicating the presence of Salmonella in the quahaug, Mercenaria mercenaria. J. Milk Food Tech. 39:322.

2. Andrews, W. H., C. D. Diggs, M. W. Presnell, J. J. Miescier, C. R. Wilson, C. P. Goodwin, W. N. Adams, S. A. Furfair, and J. F. Musselman. 1975. Comparative validity of members of the total coliform and fecal coliform groups for indicating the presence of Salmonella in the eastern oyster, Crassostrea virginica. J. Milk Food Tech. 38:453.

3. Centers for Disease Control. 1981. Annual foodborne disease summary, 1979. Centers for Disease Control, Atlanta.

4. Centers for Disease Control. 1981. Salmonella surveillance annual summary 1979. Centers for Disease Control, Atlanta.

5. Centers for Disease Control. 1980. Morbidity and mortality weekly report, annual summary. 1979. Centers for Disease Control, Atlanta.

6. Colwell, R. R., J. Kaper, and S. W. Joseph. 1977. Vibrio cholerae Vibrio parahaemolyticus and other vibrios: Occurrence and distribution in Chesapeake Bay. Science 198:394.

7. D'Aoust, J. Y., P. Stotland, and A. Boville. 1982. Sampling method for detection of Salmonella in raw chicken carcasses. J. Food Sci. 47:1643.

8. D'Aoust, J. Y., R. Gelinas, and C. Maishment. 1980. Presence of indicator organisms and recovery of Salmonella in fish and shellfish. J. Food Prot. 43:679.

9. Hendricks, C. W. 1971. Increased recovery rate of salmonellae from stream bottom versus surface waters. Appl. Microbiol. 21:379.

10. Hood, M. A., G. E. Ness, and G. Rodrick. 1981. Isolation of Vibrio cholerae serotype 01 from the eastern oyster, Crassostrea virginica. Appl. Environ. Microbiol. 41:559.

11. Hunt, D. A. 1977. Indicators of quality for shellfish water, pp. 337-345. In A. W. Hoadley and B. J. Dutka (eds.), Bacterial indicators--health hazards associated with water. American Society for Testing and Materials, Philadelphia.

12. Hussong, D., R. R. Colwell, and R. M. Weiner. 1981. Seasonal concentration of coliform bacteria by Crassostrea virginica, the eastern oyster, in the Chesapeake Bay. J. Food Prot. 44:201.

13. Janssen, W. A. 1974. Oysters: Retention and excretion of three types of human waterborne disease bacteria. Health Lab. Sci. 11:20.

14. Kelly, C. B. and W. Arcisz. 1954. Survival of enteric organisms in shellfish. Pub. Health Reports 69:1205.

15. National Marine Fisheries Service. 1982. Fisheries in the United States. 1981. National Marine Fisheries Service, United States Department of Commerce, Washington, D.C.

16. Oblinger, J. L. and J. A. Koburger. 1974. Understanding and teaching the most probable number technique. J. Milk Food Tech. 38:540.

17. Slanetz, L. W., C. H. Bartley, and K. W. Stanley. 1968. Coliforms, fecal streptococci and Salmonella in seawater and shellfish. Health Lab. Sci. 5:66.

18. Thi Son, N. and C. H. Fleet. 1980. Behavior of pathogenic bacteria in the oyster, Crassostrea commercialis, during depuration, re-laying and storage. Appl. Environ. Microbiol. 40:994.

19. United States Food and Drug Administration. 1978. Bacteriological analytical manual, 5th edition, Chapter VI, pp. 1-29. Asssociation of Official Analytical Chemists, Washington, D.C.

# ISOLATION OF INDOLE-PRODUCING BACTERIA FROM WHITE SHRIMP, (PENAEUS SETIFERUS)

R.L. Smith, R. Nickelson and G. Finne
Seafood Technology Section
Animal Science Department
Texas A&M University
College Station, Texas 77843

## INTRODUCTION

During 1979 a series of shrimp decomposition workshops were sponsored by the U.S. Food and Drug Administration, the National Marine Service and the National Fisheries Institute. The primary purpose of the workshops was to train industry personnel to recognize the typical Class 1, 2 and 3 decomposition used by the FDA to evaluate raw shrimp. Shrimp (whites, pinks and browns) were "laboratory decomposed" at various temperatures until they fell into the respective classes. The shrimp were frozen and subsequently evaluated by the workshop participants. All participants in the three workshops agreed on the classification of the shrimp which were confirmed by indole levels determined by FDA. Although there was clear agreement on the classification of the shrimp, there was a general consensus that the odors were not typical of shrimp that had undergone spoilage at refrigeration temperatures. The purposes of this study were (i) to determine if the production of indole was due to autolytic enzymes, bacterical action or a combination of the two; (ii) to isolate indole-producing bacteria from shrimp decomposed at different storage temperatures, and (iii) identify the growth characteristics of the bacteria responsible for indole production.

## MATERIALS AND METHODS

The antibacterial compounds chloramphenicol, dihydrostreptomycin sulfate, procaine penicillin, and thimerosol were added to shrimp homogenate stored at 22, 12, and 4°C for 1, 2 and 4 days respectively. Aerobic plate counts and indole levels were determined at 6 hr intervals.

Shrimp were divided into six lots, each of which was homogenized heads-on in the ratio 2 parts sterile, distilled water: 1 part shrimp. L-tryptophan was added to 3 of the lots at a concentration of 10mg/100g shrimp. Lots were paired and stored at 22, 12, and 4°C until Class 3 spoilage. Samples were taken at 3 hr intervals for the 22°C lots and 8 hr intervals for the 12 and 4°C lots then plated by spreading onto trypticase soy agar (TSA). Plates were incubated at 22°C for 48 hr for samples from all homogenates and at 12°C for 4 days and 4°C for 7 days for samples from lots stored at those temperatures. Individual

colony types were isolated and tested for indole production by the method of Vracko and Sherris (5). Indole-positive organisms were identified to the genus and when possible to the species level (1,2,3,4).

Standard Methods Caseinate Agar (SMCA) was prepared according to the method of the American Public Health Association (2) using standard methods agar (Baltimore Biological Laboratories) and sodium caseinate, sodium citrate, and calcium chloride (Fisher Scientific Co.). SMCA plates were inoculated with the indole-positive isolates, incubated at the original temperature of isolation, and examined for precipitation of clearing of the medium, both indicative of proteolytic activity.

A sterile shrimp extract was prepared by homogenizing 2 parts sterile water with 1 part shrimp; centrifuging at 20,000 x g for 1 hr at 10°C; then filter-sterilizing the supernatant through a Gelman GA-6 0.45 um membrane (Gelman Instrument Co.). Indole-positive isolates were inoculated into 0.5 ml sterile shrimp extract and 0.5 ml extract containing 2.5 mg L-tryptophan and incubated at the original temperature of isolation.

## RESULTS AND DISCUSSION

Indole production was reduced in homogenates containing chloramphenicol, dihydrostreptomycin sulfate and procaine penicillin. Bacterial growth was retarded but not eliminated. Indole production was not evident in homogenate treated with thimerosal where bacterial growth was inhibited. These results indicate indole production is due to bacterial tryptophanase and not endogenous shrimp enyzmes.

One thousand six hundred and forty seven isolates were selected for indole production testing. Only 42 (2.6%) of the isolates were positive. Of the organisms identified as indole producers, 52% were Flavobacterium, 24% Aeromonas and 21% Proteus. No Escherichia coli were isolated.

In evaluating growth characteristics it was noted that the Aeromonas and Proteus isolates only appeared at the 22°C incubation, whereas all isolates capable of producing indole as low as 2°C belonged to the genus Flavobacterium. Aeromonas and Proteus were also proteolytic on SMCA while Flavobacterium was not. The ability to hydrolyze protein and produce indole were closely related in an isolate's ability to produce indole in sterile shrimp extract. Only 16 isolates were positive in the extract without L-tryptophane (all Aeromonas and Proteus), whereas 36 isolates were positive when L-tryptophane was added.

## CONCLUSION

Indole is the product of bacterial tryptophanase in shrimp. There appears to be two routes by which indole is produced. First is the typical high temperature abuse associated with Class 1, 2 and 3 decomposition. The organisms responsible are mesophilic and proteolytic and belong to the genera Aeromonas and Proteus. The second route is low temperature and more typical of poor quality shrimp after prolonged storage

at refrigeration temperature. The organisms responsible are psychrotrophic, non-proteolytic (produce indole only after proteolysis by typical spoilage organisms like Pseudomonas) and belong to the genus Flavobacterium.

## REFERENCES


1. Buchanan, R.E., and N.E. Gibbons. 1974. Bergey's manual of determinative bacteriology, 8th ed. Williams and Wilkins Co., Baltimore, Md. USA. 1246 p.

2. Intersociety/Agency Committee on methods for the biological examination of foods. 1976. Compendium of methods for the microbiological examination of foods. American Public Health Association, Washington, D.C.

3. Lewis, D.H. 1973. Predominant aerobic bacteria of fish and shellfish. Texas A&M University Sea Grant Publ. No. 73-401.

4. Vanderzant, C., and R. Nickelson. 1969. A microbiological examination of muscle tissue of beef, pork, and lamb carcasses. J. Milk Food Technol. 32:357-361.

5. Vracko, R., and J. C. Sherris. 1963. Indole-spot test in bacteriology. Am. J. Clin. Path. 39:429-432.

# "ON THE RELATIONSHIP BETWEEN $T_bOD$ AND COD IN INDUSTRIAL LIQUID EFFLUENTS FROM FISH PROCESSING PLANTS"

María A. Parín, Estela M. Civit
Centro de Investigaciones de Tecnología Pesquera (CITEP)- INTI
Marcelo T. de Alvear 1168 - 7600 Mar del Plata - Rep.Argentina
Héctor M. Lupín
Fish Utilization and Marketing Service Fisheries Department, FAO
Via delle Terme di Caracalla - 00100 Rome - Italy

## ABSTRACT

Treatment of liquid effluents from the fishing industry has become a matter of interest to many countries. Since these effluents usually contain large quantities of organic matter, they present special problems for the planning and operation of biological treatments.

This paper deals with the evaluation of the biodegradable organic matter present in such effluents. The method proposed by Mullis and Schroeder (1971) to determine the total biochemical oxygen demand ($T_bOD$) was applied to effluents from fish filleting and fish meal plants. A relationship was found between $T_bOD$ and the chemical oxygen demand (COD). The difficulties in applying the methods are also discussed.

## INTRODUCTION

The fishing industry usually produces a large quantity of liquid waste. The amount of liquid waste and its composition depend on the processing method used (10). Composition of waste also depends on the raw materials (13).

Effluents from fish processing plants contain large quantities of proteinaceous and fatty compounds.As a result, the charge of a fishing effluent might be very high when compared with common sewage and effluents from other food industries, with the possible exception of those from slaughterhouses. The difficulties that researchers and the fishing industry experience when trying to apply standard tests to fishing waste effluents are well known. These difficulties are very often associated with the unusually high concentration of organic matter. The problem is not only related to the choice of the most suitable method to monitor the effluents, but also to the design and proper application of biological treatments.

The most widely used test to assess the organic pollutant is the Biochemical Oxygen Demand (BOD), based on the bacterial oxidation of the organic matter present in the effluent. The test is carried out at 20°C for five days (1), (2) and (16). Effluents with a high organic charge need high dilutions since the oxygen solubility in water is very low (9.17 ppm at 20°C) but the higher the dilution the lower the precision.

Another problem that affects BOD when applied to fish effluents (or effluents other than common sewage) is that the 5- day period might not be representative. If micro-organisms are not in their log phase when the test begins the BOD curve will show a delay which may influence the reading on

the fifth day (6), (7) and (15).

Another widely used test to measure pollutants is the Chemical Oxygen Demand (COD) based on the oxidation of the organic matter present in the effluent by potassium dichromate. COD can be measured in only two to three hours (1). The time factor becomes important when plants are being monitored.

Other quick instrumental methods have also been developed to measure pollutant organic charge, such as the Total Organic Carbon (TOC) and the Total Oxygen Demand (TOD). Although TOC and TOD values can be readily obtained in a few minutes, they call for a high initial cost in instruments (10).

The real problem is that COD, TOC and TOD methods fail to give a clear indication of the biodegradability of the organic substances present in the effluent (16).

To overcome some of the problems the BOD and COD test present, Busch (3) Busch et al. (4) and Hiser & Busch (9) proposed the Total Biochemical Oxygen Demand test ($T_bOD$). The $T_bOD$ test uses the COD test and a mass culture of microbial cells. The $T_bOD$ test can be used to measure the change in COD caused by biological oxidation. This change in COD would be equal to the equivalent biological oxygen demand of the liquid effluent.

The $T_bOD$ test is not directly applicable to waste effluents containing insoluble matter since biodegradable, particulate organic matter cannot be distinguished from microbial cells.

Mullis and Schroeder (11) studied domestic sewage and proposed expanding the $T_bOD$ test to systems containing a significant amount of insoluble organic material. The basic assumption of Mullis and Schroeder (11) was that all incoming solids are oxidized. They also found an empirical linear correlation between $T_bOD$ and COD in domestic sewage and suggested to run the $T_bOD$ test intermittently throughout the year to maintain the validity of the relationship for operational purposes. In daily plant operations, the COD test may be sufficient to ascertain biodegradable matter, not by its actual value but its relationship with $T_bOD$.

Liquid effluents from fish processing plants usually contain large portions of insoluble organic matter and have a higher organic concentration than domestic sewage. This paper deals with application of the $T_bOD$ test to the liquid effluents of fish industries and the relationship between $T_bOD$ and COD. The effluents tested were bloodwater, an effluent derived from fish meal plants, and the liquid effluent from whiting filleting plants.

## MATERIALS AND METHODS

Integrated samples of bloodwater were collected from the daily drainage after decantation of the pits, in fish meal factories in Mar del Plata. Integrated samples of liquid wastes from filleting of whiting were also taken from the drainage after decantation. Samples were transported to the laboratory in sealed flasks refrigerated with crushed ice.

The average composition of fish bloodwater and fish filleting wastes have already been published (13), (5) and (8).

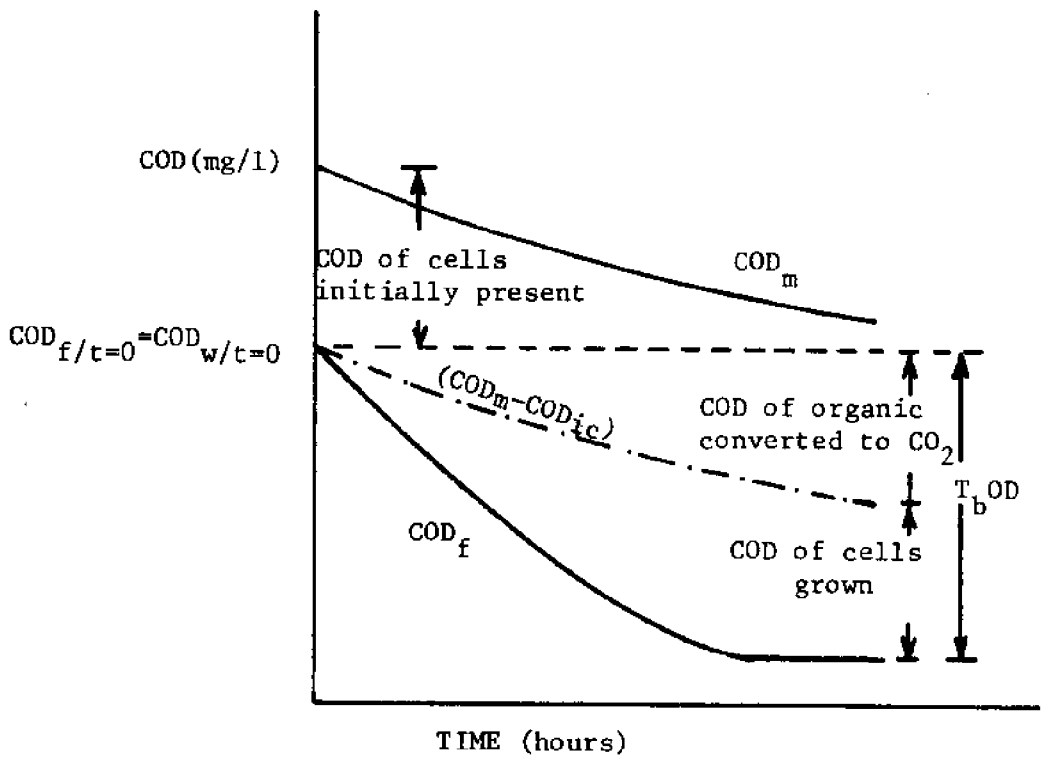
The Chemical Oxygen Demand (COD) was measured according to APHA (1).

The basic procedure to follow for the $T_bOD$ test is described below (16):

1) An acclimated and washed cell suspension is obtained.
2) COD and mass concentration are determined on the cell suspension.
3) COD of the waste water is determined.
4) The acclimated cell suspension and the waste water are mixed in predetermined proportions.
5) The mixture is aerated using a diffuser stone.
6) Samples are taken and analysed for total (mixture) COD, filtrate (0,45 μm) COD, and suspended solids at convenient time intervals.
7) Water is added as needed to replace evaporation.

By plotting the results of COD vs time, it is possible to obtain the ultimate waste water biochemical oxygen demand ($BOD_L$), the residual COD after treatment and the estimated quantity of oxygen (or air) necessary for the treatment under experimental conditions (see Figure 1).

FIGURA 1: Typical $T_bOD$ test results for wastewater not containing organic insoluble matter.



The acclimated cell suspension was obtained by using the same effluent as a microbiological medium. The followed procedure is:

1) A sample of liquid effluent from fish processing plants is filtered using Whatman paper N° 42. Bloodwater is diluted by 50%.

2) 300 ml of the filtered waste water are mixed with 1,500 ml of bidistilled water enriched with nutrients (1), 20 ml of buffer solution of phosphate (1) and 2 ml of $ClNH_4$, 1M.

3) The mixture is aerated using a diffuser stone for 24 hours at room temperature.

4) After aeration, the mixture is filtered through Whatman GP filter paper. The mass of cells is washed twice by centrifugation and diluted in tap water with nutrients to 1,250 ml. Then, the culture is ready to be used.

In the original method (9) for effluents without suspended matter, the sample to be analysed is also filtered through Whatman GP. The filtrate is mixed with the measured proportions of the acclimated cell suspension. The COD in the mixture is then determined ($COD_m$) using the following expression:

$$COD_m = \frac{V_c \ COD_c + V_e \ COD_e}{V_c + V_e}$$

where:

$V_c$ = volume of acclimated cell suspension.

$V_e$ = volume of effluent.

$COD_c$ = chemical oxygen demand of the cell suspension.

$COD_e$ = chemical oxygen demand of the effluent.

The COD of the filtrate passing a 0.45 µm filter is also measured ($COD_f$).

In the case of waste water containing insoluble matter, the $T_bOD$ test cannot be applied in the same way. Biodegradable suspended organic matter cannot be differentiated from cells. Although this applies to the $COD_f$ curve in Figure 2, since non-filterable organics are not measured, only the final plateau can be used to determine the $T_bOD$.

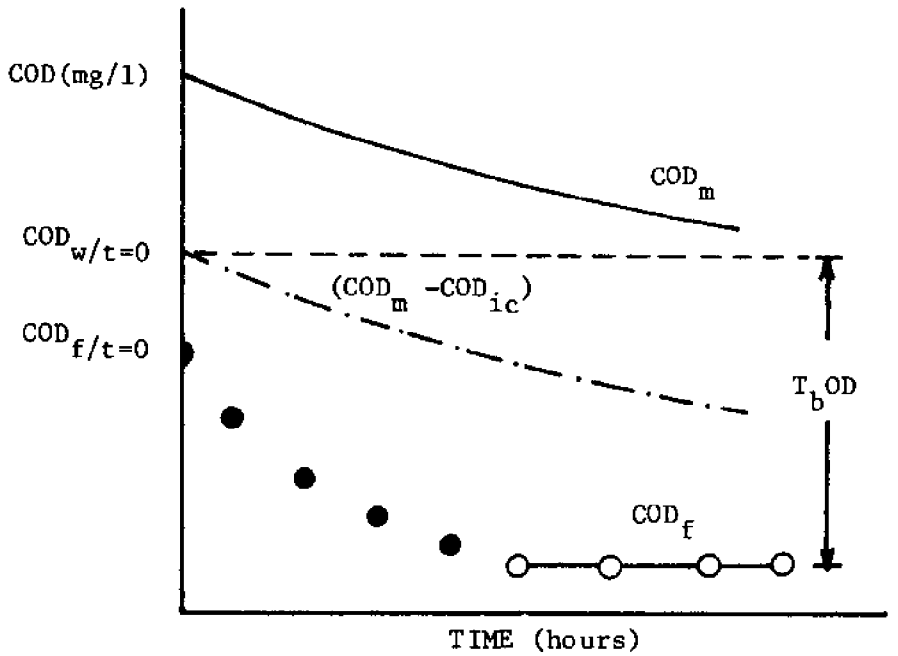
It is clear from Figure 1 that the initial point of both $COD_f$ and ($COD_m$ - $COD_{ic}$) curves is the initial COD of the waste water to be tested ($COD_{w/t=0}$), which is obviously influenced by the dilution factor. This value can be readily determined in the waste water with suspended organic matter. But, unlike the case shown in Figure 1, for waste waters containing suspended organic matter,

$$COD_{f/t=0} \neq COD_{w/t=0}$$

However, the end point, when the value of $COD_f$ remains stationary, is an indication that the bio-oxidation process is complete and it is the lower limit attainable in COD reduction. Now the (quasi) steady state reached by the $COD_f$ curve can also be used for calculating $T_bOD$.

The $COD_m$ data provide the same information as the $T_bOD$ test for soluble wastes. A typical test for waste water containing insoluble organic matter is shown in Figure 2.

FIGURE 2: Typical $T_bOD$ test results for waste water containing organic insoluble matter.




## EXPERIMENTAL RESULTS

### Bloodwater

The initial COD of the bloodwater studied ranged from 20,000 to 100,000 mg/1. In order to get satisfactory results, samples should be diluted when mixed with the acclimated cell suspension. It was found that a workable ratio is 1:5 in volume.

For practical purposes in the COD test, bloodwater should be diluted to 1 or 2%.

The COD test and the chemicals used were analysed in recuperation trials using potassiumacid phthalate ($C_8H_4O_4KH$) as reference substance. The percentages of recuperation were found to be between 98.4 and 99%.

The whole $T_bOD$ experiment was analysed in recuperation trials with glutamic acid as the reference substance. The percentages of recuperation were found to be between 90 and 106%, somewhat higher limits than those found by Hiser y Busch (9) when testing the method for waste water not containing organic matter in suspension (96 to 103%).

Preliminary COD test have shown that the heating time during the experiment should, in this case, be 90 minutes.

The variation in the value of mixed liquor suspended solids (MLSS), i.e. the solids retained weight when filtering through 0.45 μm, is of no significance when analysing waste water containing suspended organic matter. Nevertheless, the MLSS of the acclimated cell suspension and the $T_bOD$ may be used to define a waste water charge. This waste water charge is useful for adjusting the test time. For glutamic acid, a charge of 0,40 $T_bOD$ (mg/l)/MLSS (mg/l) needs around 5 hours for reaching the plateau while higher charges need longer times (14). On the other hand, a too low charge increases the risk of error due to the effect of dilution.

Figure 3 shows the results of a typical $T_bOD$ test of bloodwater. The relationship between the $T_bOD$ and the COD of the waste is shown in Figure 4.

FIGURE 3: Typical $T_bOD$ test results for bloodwater.

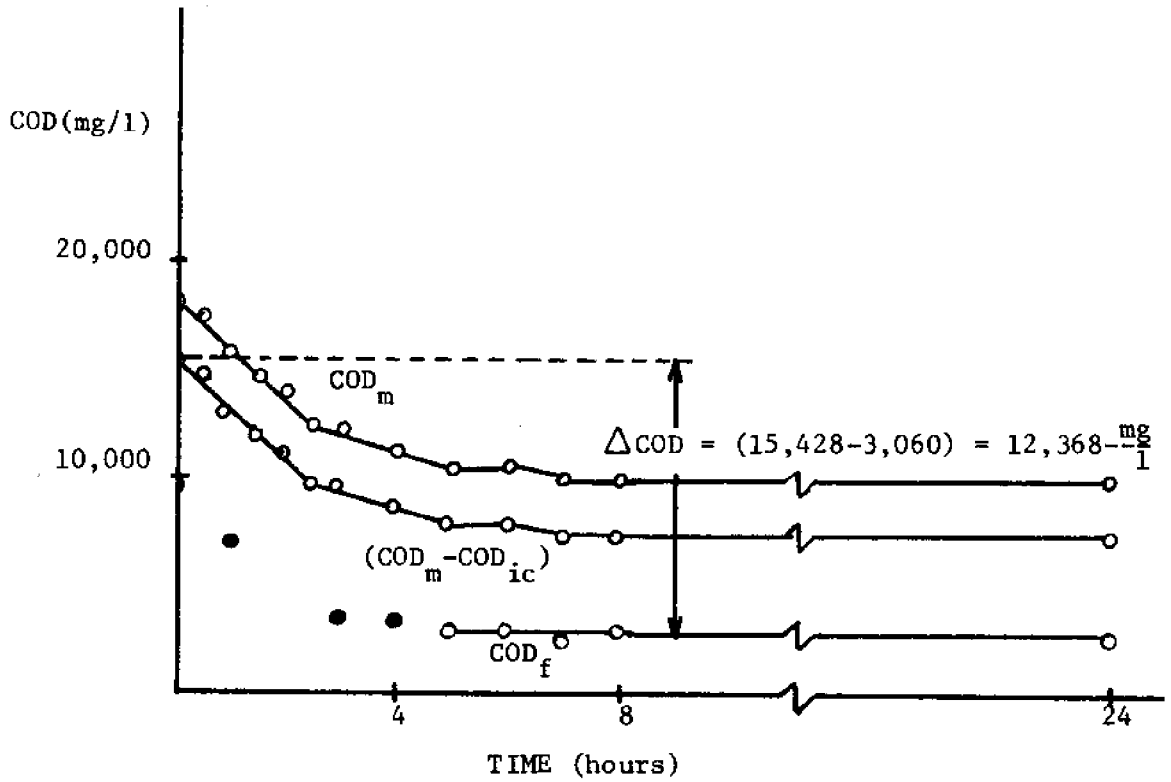
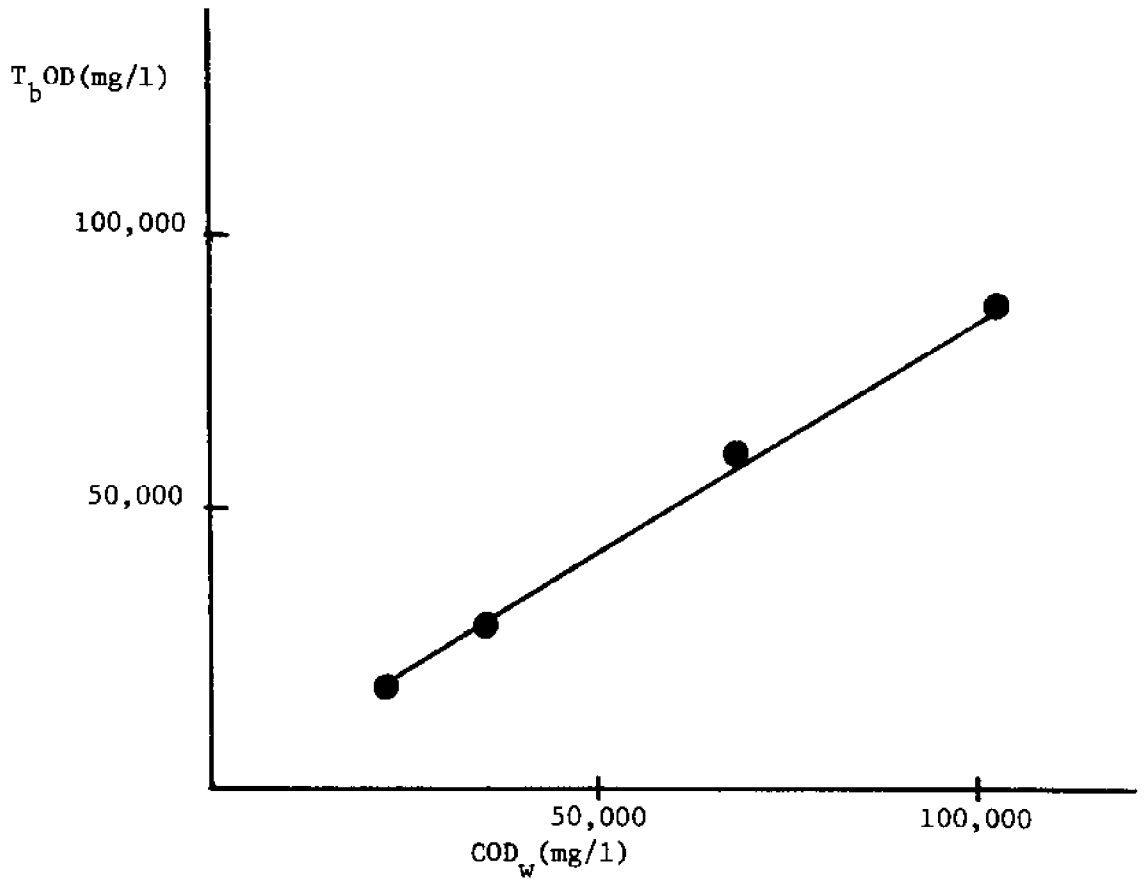


FIGURE 4: $T_bOD$ versus $COD_w$ for bloodwater
$T_bOD = 0.851\ COD_w - 1306.967$ (r = 0.999)




Filleting waste water

The initial COD of the filleting waste water studied ranged from 400 mg/l to 1.000 mg/l. Although this is considerably lower than the bloodwater range, it is higher than the range of common sewage (50 - 300 mg/l).

It was not found necessary to dilute the samples when mixing them with the acclimated cell suspension. The ratio of waste water to acclimated cell suspension was 9:1. The other conditions were similar to those of the $T_bOD$ experiment for bloodwater as described above.

Figure 5 shows the results of a typical $T_bOD$ test for filleting waste water. Figure 6 shows the relationship between the $T_bOD$ result and the initial COD of the filleting waste water.

## DISCUSSION

The $T_bOD$ test can be applied to waste waters from fish processing plants. A correlation was developed between the $T_bOD$ and COD of the waste waters.

FIGURE 5: Typical $T_bOD$ test results for filleting waste water

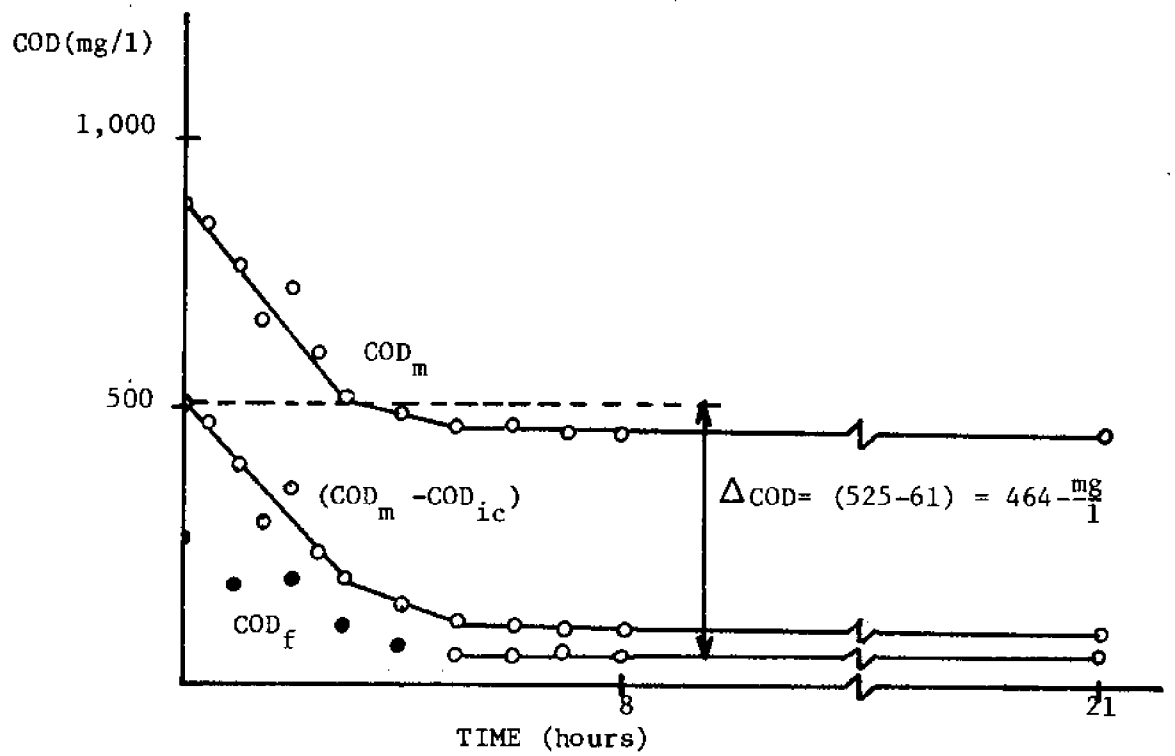
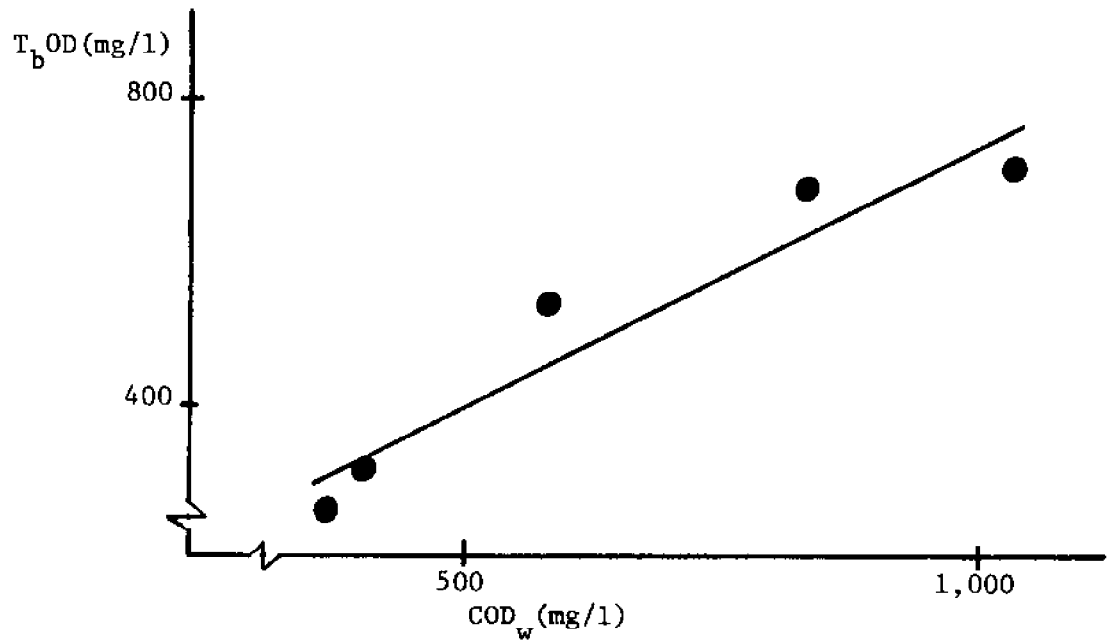



FIGURE 6: $T_bOD$ versus COD for filleting waste water
$T_bOD = 0,687\ COD_w + 50,158\ (r = 0,954)$



Although the relationship between $T_bOD$ and COD is quite useful, it should be borne in mind that it is an empirical correlation which should be periodically checked for each type of effluent and situation.

The relationship between $T_bOD$ and $COD_w$ should be closely studied during each treatment and at the same time the correlation between both parameters could be improved.

From the $T_bOD$ test can be calculated the quantity of oxygen necessary for the treatment (it depends on the culture used) and the residual COD after treatment. These values for bloodwater and fish filleting waste water are shown in Table 1.

TABLE 1.

| FISH WASTE WATER | | OXYGEN UPTAKE (mg/l) | RESIDUAL COD (mg/l) |
|---|---|---|---|
| BLOODWATER | A1 | ---- | 17,500 |
| | A2 | ---- | 9,780 |
| | A3 | 11,763 | 4,748 |
| | A4 | 15,300 | 7,489 |
| FILLETING | F1 | 242 | 112 |
| | F2 | 467 | 67 |
| | F3 | 283 | 95 |
| | F4 | 683 | 343 |
| | F5 | 516 | 146 |

The COD test can be easily implemented and it is not expensive. The whole experiment can be completed in less than two hours. The whole $T_bOD$ test is also relatively easy to carry out.

At very high values of $COD_w$ (above 50.000 mg/l), the difference between $COD_w$ and $T_bOD$ becomes minimal and falls within the experimental error expected for this kind of test. Further studies seem necessary to analyse the application of the test results in this case.

## ACKNOWLEDGEMENTS

This work was supported in part by the Consejo Nacional de Investigaciones Científicas y Técnicas and the Comisión de Investigaciones Científicas de la Provincia de Buenos Aires of Argentina. The authors are indebted to Arpemar S.A., Ventura S.A. and Pesquera Roda, fish processing industries of Mar del Plata, for their collaboration.

## REFERENCES

(1) American Public Health Association (APHA). "Standard Methods for the Examination of Water and Wastewaters". 14 th Ed., Washington, DC (1975).

(2) Bailey, J.E. and Ollis, D.F.

"Biochemical engineering fundamentals."
Ed. McGraw-Hill, 1977.

(3) Busch, A.W.
"B.O.D. progression in soluble substrates".
Proceedings 13th, Ind. Waste Conf., Purdue Univ., 1958.

(4) Busch, A.W.; Grady, L. Jr.; Shivaji Rao, T. and Swilley, E.L.
"Short-term total oxygen demand test".
Journal of Water Pollution Control Federation (J.W.P.C.F.), 34:4, 1962.

(5) Civit, E.M.; Parín M.A. y Lupín, H.M.
"Recovery of protein and oil from fishery bloodwater waste. Effect of pH and temperature".
Water Res. Vol. 16, pp 809 to 814 (1982)

(6) Flegal, T.M. and Schroeder, E.D.
"Temperature effects on BOD stoichiometry and oxygen uptake rate".
J.W.P.C.F., 48: 12, 1976.

(7) Gaudy, A.F. Jr.; Bhatla, M.N.; Follet, R.H. and Abu-Niaaj, F.
"Factors affecting the existence of the plateau during the exertion of BOD".
J.W.P.C.F., 37: 444, 1965.

(8) Gonzalez, J.F.; Civit, E.M. y Lupín, H.M.
"Composition of fish filleting waste-water (in press).

(9) Hiser, L.K. and Busch, A.W.
"An 8-hour biological oxygen demand test using mass culture aeration and COD".
J.W.P.C.F., 36: 505, 1964.

(10) Kehoe, T.J.
"BOD, COD and TOC. After ten years of talk"
Pollution Engineering, p 49, 1980.

(11) Mullis, M.K. and Schroeder, E.D.
"Use of the $T_b$OD test with colloidal wastewaters".
Proc. 25th, Ind. Waste Conf., Purdue, Univ., 1970.

(12) Nemerow, Nelson.
"Industrial Water Pollution. Origins, characteristics and treatment".
Ed. Addison-Wesley, 1978.

(13) Parín, M.A.; Civit, E.M. y Lupín, H.M.
"Characterization of bloodwater effluent of fish meal factories"
Latin Am. J. Chem. Engng. Appl. Chem. 9, 155-164 (1979)

(14) Parín, M.A.
Informe Beca Perfeccionamiento de CONICET (1981)

(15) Schroeder, E.D.
"Importance of the BOD plateau".
Water Research, 2: 803-809, 1968.

(16) Schroeder, E.D.
"Water and wastewater treatment".
Ed. McGraw-Hill, 1977.

# CLOSED SYSTEM SHEDDING OF BLUE CRABS: THE VIRGINIA EXPERIENCE

Michael J. Oesterling and William D. DuPaul
Marine Advisory Services
College of William and Mary
Virginia Institute of Marine Science
Gloucester Point, Virginia

## ABSTRACT

Virginia has consistently been a leader in the production of soft shell crabs. For the period 1977-1981, the average production of soft crabs has exceeded 317,520 kg, valued at over $735,000.

Traditionally, this production has been achieved using methods little changed from the earliest beginnings of the industry. The use of in-water floating boxes (also known as "floats") is still an important means of shedding crabs. During the 1950's on-shore flow-through shedding tanks were introduced. Today, many producers have switched entirely to on-shore shedding, or rely on a combination of floats and tanks. Over the past several years a great deal of interest has been expressed regarding the shedding of crabs in closed, recirculating water systems.

In Virginia, the closed systems used vary in design and complexity. Currently, there are 3 basic facility designs in production. Each of these will be briefly discussed as to construction and advantages or disadvantages.

# CLOSED SYSTEM SHEDDING BLUE CRABS: THE VIRGINIA EXPERIENCE

Michael J. Oesterling and William D. DuPaul
Marine Advisory Services
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## INTRODUCTION

The blue crab (Callinectes sapidus Rathbun) supports a large commercial fishery along the eastern seaboard of the United States and Gulf of Mexico (7). In actuality there are 2 blue crab fisheries - one for hard-shelled and one for soft-shelled crabs. Soft-shelled crabs are not a separate species of crab, but are blue crabs that have shed (molted) their hard outer shells in preparation for growth. Virginia has consistently been a leader in the production of soft-shelled crabs. For the period 1977-1981, the average production of soft crabs in Virginia has exceeded 317,520 kg, valued at over $735,000 (P.J. Anninos, Virginia Marine Resources Commission, personal communication).

Traditionally, this production has been achieved using methods little changed from the earliest beginnings of the industry. The use of in-water floating boxes (also known as "floats") is still an important means of shedding crabs. During the 1950's, on-shore flow-through shedding tanks were introduced. Today, many producers have switched entirely to on-shore flow-through shedding, or rely on a combination of floats and tanks.

In spite of the information accumulated in over 125 years of shedding soft crabs, producers at times encounter excessive mortalities of crabs during the molting process. Many of these mortalities can be traced to poor water quality and, in some cases, are site specific (1). Lately, interest has been expressed in the use of a water recirculating, or closed, system for crab shedding as a means of better controlling water quality (2, 3, 4, 5). In areas of poor water quality, the recirculating system offers a viable method for shedding soft blue crabs. The remainder of this paper will deal with the application and design of closed system blue crab shedding by Virginia producers.

## DISCUSSION

In Virginia, the closed systems being used vary in design and complexity. However, the basic principles of closed, recirculating

seawater systems as discussed by Spotte (6) apply. Operators have adapted this information to suit their own needs. Currently, there are 3 basic facility designs in successful production in Virginia. In any one system a combination of foam fractionation, biological, mechanical or algal filtration may be found.

The first system, and the simplest, is essentially a large, shallow aquarium (Figure 1). Only one individual is currently employing this method in Virginia. This method does not pump water, but uses an air-lift system to recirculate water through a filter bed. A 1.22 m by 2.44 m tank is used, constructed with a filter in the tank itself. A false-bottom of peg-board or stainless steel wire is supported 5 cm above the bottom of the tank creating a space filled with water. On top of the false-bottom is approximately 5 cm of crushed oyster shell forming the basis for a biological filter unit. Over the crushed shell is 10 to 15 cm of water. At the 4 corners of the tank are 3.8 cm diameter polyvinyl chloride (PVC) pipes that extend below the false-bottom and 2 to 5 cm above the water level. In each pipe is an aquarium-type air stone and plastic tubing connecting the air stone to a main air delivery line, also of plastic. A 1/3 horsepower air compressor is used to supply air to 8 tanks within the entire system.

The production capabilities of this system have yet to be adequately assessed. However, there does seem to be potential for large-scale production of soft crabs using this type of closed system.

The remaining 2 closed systems are similiar to each other. They both pump water and incorporate biological filtration, foam fractionation and mechanical filtration.

Located on Virginia's Eastern Shore, the first facility has twenty, 1.22 by 2.44 m wooden tanks, situated outside without protection from the elements. A foam fractionation unit, or protein skimmer, is an integral part of this closed system. It is necessary to elevate the skimmer to create enough head-pressure to permit gravity distribution of water to the shedding tanks. Some operators not wanting the added expense of a telephone pole have used more resourceful methods to mount their skimmers, including using a nearby tree. The skimmer itself is quite easily constructed (Figure 2). It consists of a 3 to 4.5 m long piece of PVC pipe with a diameter of 15.25 to 30.5 cm. Connected to the main pipe is an input from a biological filter and an out-flow to the shedding tanks. These pipes are 3.5 to 5 cm diameter PVC pipe. The placement of these pipes is important to proper functioning of the skimmer. The input pipe extends up the outside of the main pipe to within 20 to 30 cm of the top, makes two 90° angles (elbows) and then extends downward to within 15 to 20 cm of the bottom, at which point it enters the main pipe. Critical to the working of this skimmer is the placement of an aspirator at the start of the downward turn of the input line (Figure 2). When the input line enters the main pipe it is upturned and can be capped with a shower head nozzle or pinched somewhat (Figure 3). The purpose

of the nozzle head is to break-up air which was sucked in by the aspirator, thus creating small bubbles for the adsorption of proteins and the creation of foam. Many commercially available protein skimmers require either elaborate tooling of venturi devices or supplemental air supplies. However, this particular construction and aspirator negates the use of either (Figure 4). The aspirator is nothing more than a constriction in the water flow made by the insertion of a hard plastic washer-like disc at the second elbow of the input line causing a jetting action which in turn draws in air through a small hole drilled in the input pipe.

The skimmer is raised 1.8 to 3 m above the level of the shedding tanks, as previously mentioned. For the facility being described, it feeds to a main water distribution pipe (the skimmer out-flow), splits into 2 pipes and finally into 4 pipes, each serving 5 shedding tanks. All tanks then drain to a biological filter constructed from a 2840 l concrete septic tank bottom filled with oyster shells. The entire filter is buried to ground level. This facilitates gravity drainage from the shedding tanks, as well as providing some temperature moderation during the summer months. Prior to entering the filter, water passes through a basket filled with fiberglass insulation material which serves as a crude mechanical filter, catching lost crab appendages and other pieces of large debris. As the need arises, this fiberglass is replaced with clean material. The system just described is all powered by a single 2 horsepower centrifugal water pump.

The final system is similiar, but more elaborate. It has eighteen shedding tanks constructed of concrete and housed in a building. What makes this system different from the previously described system is the inclusion of an additional biological filter and another pump. There is still a protein skimmer, located on the outside of the building. The biological filters, however, are unique.

One biological filter is again constructed of a 2840 l septic tank, but the cover is translucent fiberglass permitting sunlight to pass through. Inside this filter is a trough that evenly distributes incoming drain water to a tray of whole oyster shells. There is another tray of 2.54 $cm^2$ oyster shell pieces under this tray of whole shells. At one end of the filter is a solid partition that extends to within 5 cm of the bottom. From this small compartment water is pumped to the protein skimmer. This arrangement assures that water is drawn through the filter medium. Water than passes through the skimmer and enters into the building. Here it empties into the second biological filter. Over the top of this filter is laid fiberglass insulation material. In this case the insulation material removes more dirt than anything else, since the distribution trough in the outside filter catches debris. This inside filter again uses oyster shell as a medium, but does not use any whole shell, only pieces. Again there are 2 trays of shell. The tray system was used to facilitate easy removal for cleaning, although it was not necessary

during the past production season to do so. From this filter the water is then pumped to the shedding tanks. Figure 5 details the lay-out for this system which includes: a large outside biological filter; a 1.5 horsepower pump to move water through a protein skimmer; an inside smaller biological filter with insulation material for a mechanical filter; another 1.5 horsepower pump to distribute water to the shedding tanks. There is a small problem in synchronizing the two pumps, but once done the system functions well.

## SUMMARY

Currently in Virginia there are at least 5 facilities using recirculating systems for the production of soft crabs. This number will grow as more is learned and techniques refined. The use of closed systems will make crab shedding possible for those individuals that may not have the best waterfront location, or no waterfront at all.

## REFERENCES

1. Oesterling, M. J. 1982. Mortalities in the soft crab industry: sources and solutions. Virginia Institute of Marine Science, Marine Resource Report No. 82-6, 11 pp.

2. Ogle, J. T., H. M. Perry and L. Nicholson. 1982. Closed recirculating seawater systems for holding intermolt blue crabs: literature review, systems design and construction. Gulf Coast Research Laboratory, Technical Report Series No. 3, 11 pp.

3. Paparella, M. (editor). 1979. Information tips. University of Maryland, Marine Products Laboratory, 79-3, 6 pp.

4. Paparella, M. (editor). 1982. Information tips. University of Maryland, Marine Products Laboratory, 82-2, 11 pp.

5. Perry, H. M., J. T. Ogle and L. C. Nicholson. 1982. The fishery for soft crabs with emphasis on the development of a closed recirculating seawater system for shedding crabs. Proceedings of the Blue Crab Colloquium, October 18-19, 1979, Biloxi, Mississippi, p. 137-152.

6. Spotte, S. 1979. Fish and invertebrate culture, water management in closed systems. John Wiley and Sons, Inc., New York, New York. 179 pp.

7. Williams, A. B. 1965. Marine decapod crustaceans of the Carolinas. Fishery Bulletin, 65(1):1-298.

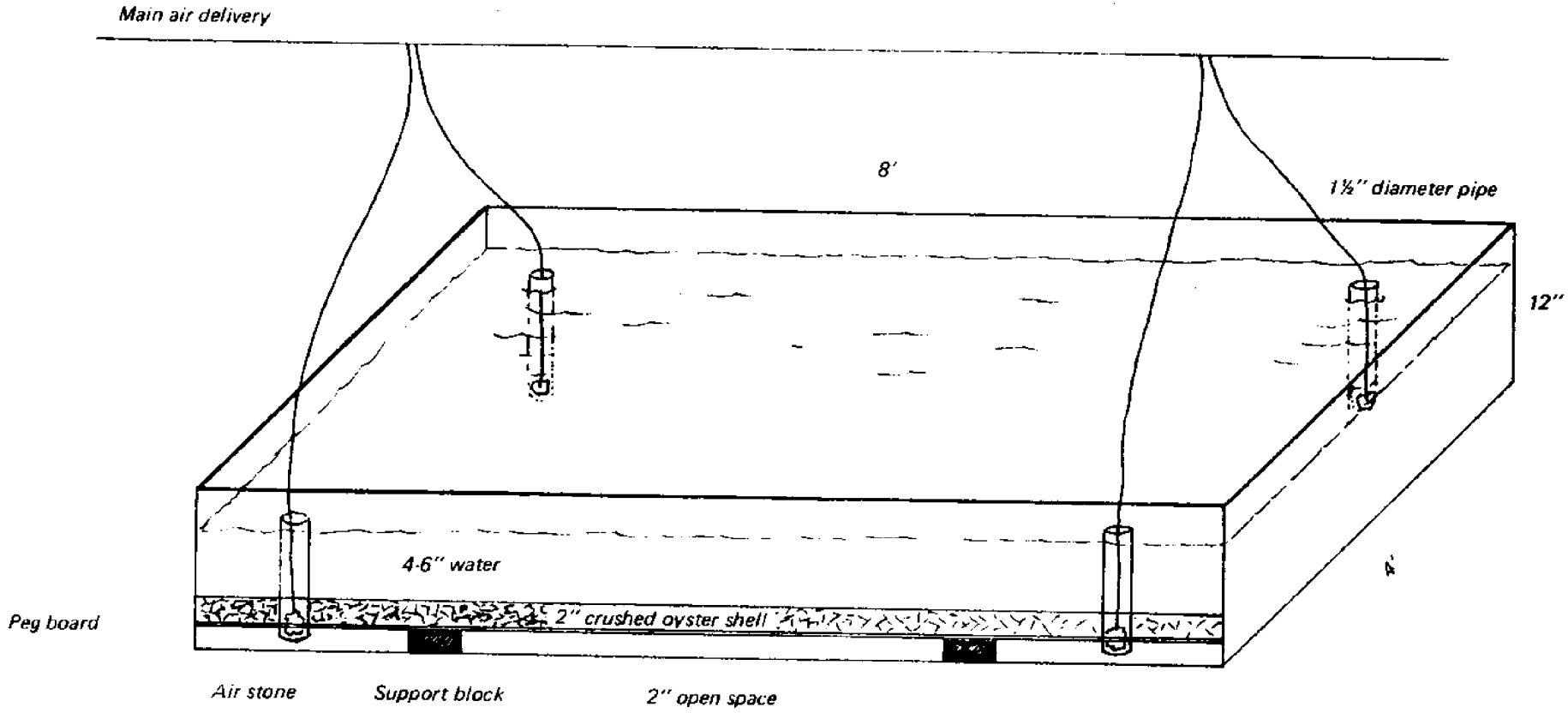



Figure 1. Aquarium-type closed system with each shedding tank containing its own biological filter. Note that an air-lift system is used to circulate water through the filter medium.

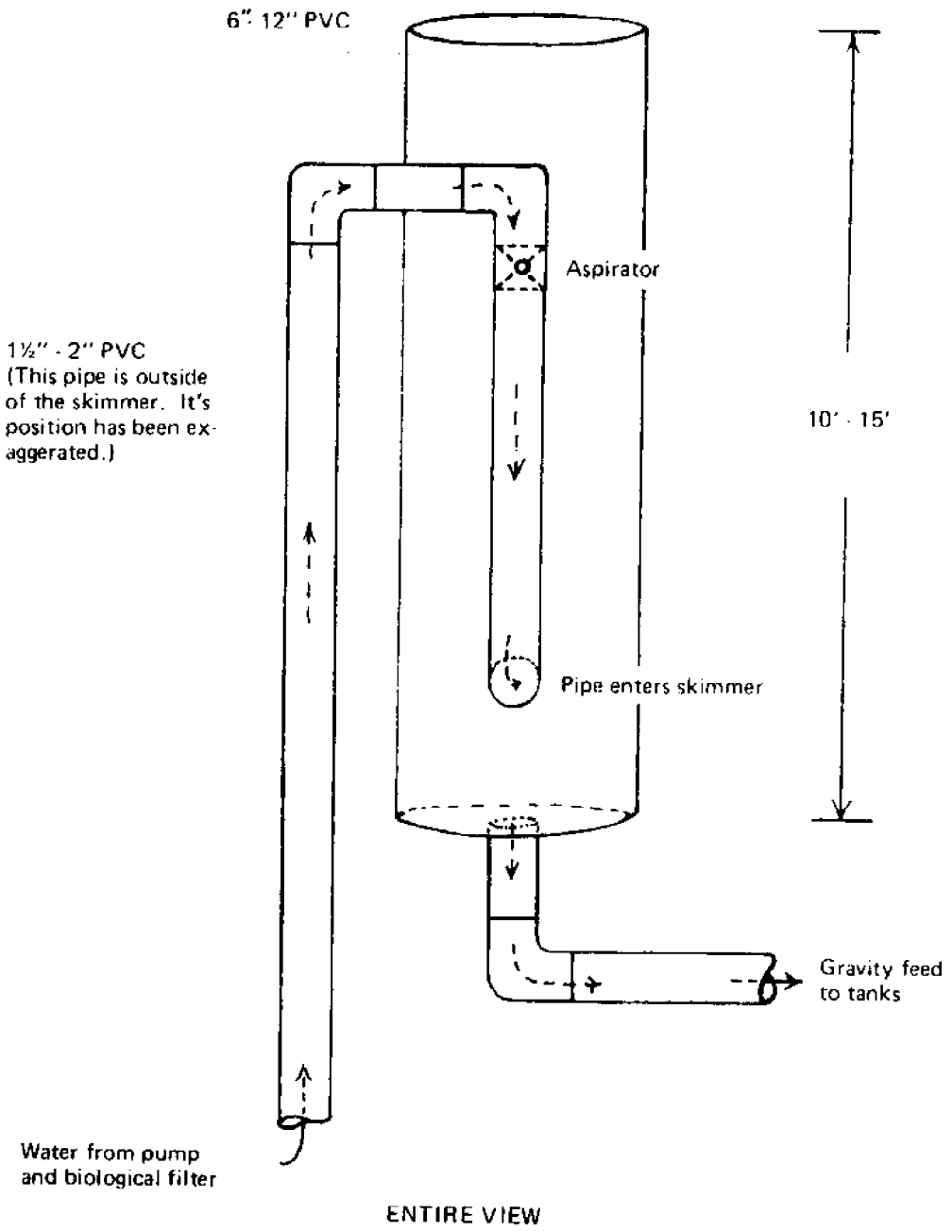



Figure 2. Foam fractionation unit used in closed system shedding of blue crabs.

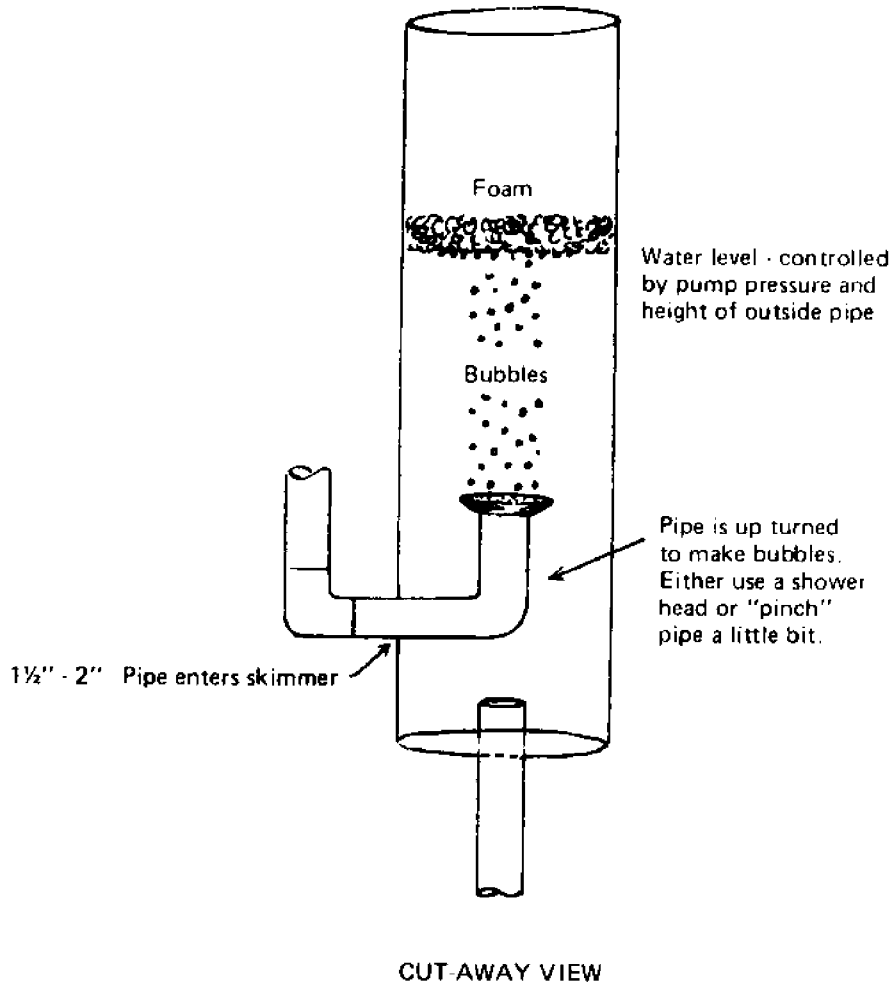
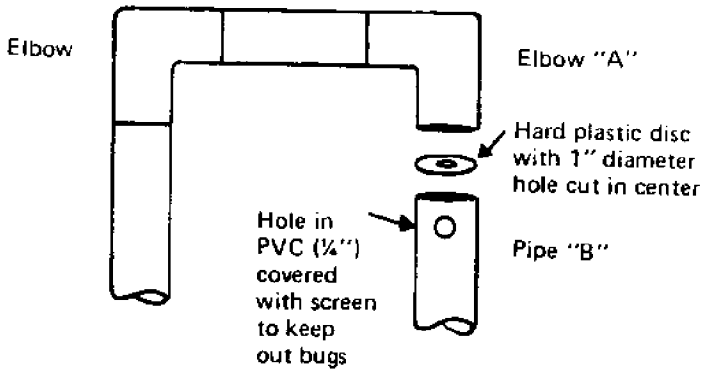



Figure 3. Foam fractionation unit used in closed system shedding of blue crabs.

ASPIRATOR CONSTRUCTION

This type of construction negates the need for fancy tooling or for an additional compressed air source




Before connecting Pipe "B" to Elbow "A" insert plastic disc. This creates a constriction, forcing water to "jet" through. This in turn sucks air in through the hole drilled in Pipe "B", oxygenating the water. The shower head (or pinched pipe) breaks up the air creating bubbles.

Figure 4. Aspirator used in foam fractionation unit in closed system shedding of blue crabs.

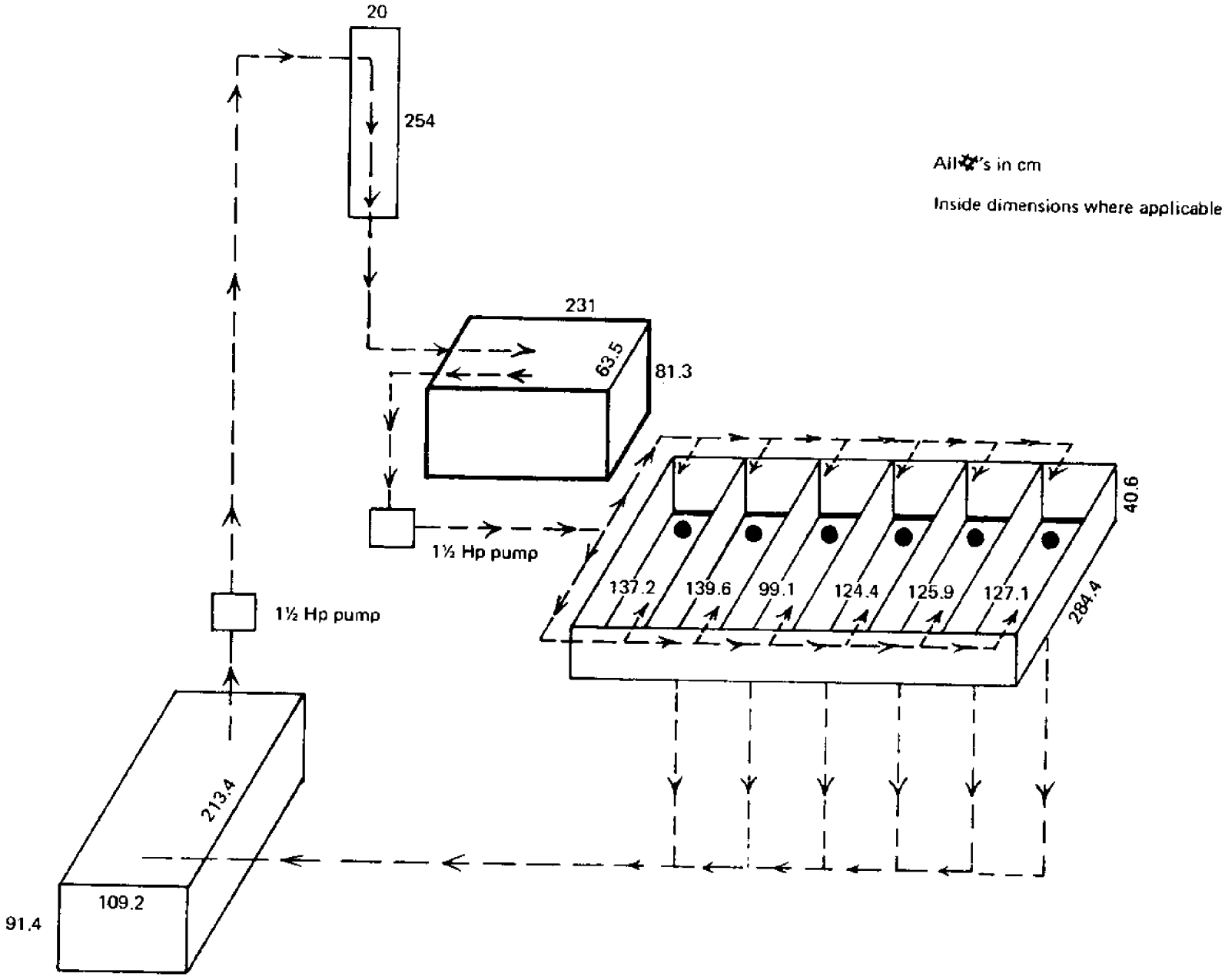



Figure 5. Schematic for a closed system blue crab shedding facility. Beginning in the lower left is a large biological filter which pumps to a protein skimmer; skimmer feeds by gravity to another biological filter; a pump distributes water from this filter to shedding tanks; gravity drains water from shedding tanks to large biological filter.

# TEXTURAL MEASUREMENTS AS AN EVALUATION OF FISH FRESHNESS

Ernest A. Johnson
Food Engineering Department
University of Massachusetts
Amherst, Massachusetts 01003

and

Ronald A. Segars and John Kapsalis
U.S. Army Natick R&D Laboratories
Natick, Massachusetts 01760

INTRODUCTION. Maintaining high quality fisheries products all the way from the fisherman through the distribution channels to the consumer has been given a high priority status by the Seafood Science and Technology Workshop in their recent identification and ranking of the most important problems in fisheries resources today( 5). The Workshop also identified an important problem in the lack of rapid methods to judge fish quality. One of the criteria for the evaluation of the quality of whole fish, included by the NMFS in their Fishery Products Inspection Manual, is the resilience or the elasticity of the fish flesh ( 3,4). This test is performed subjectively by the inspector, who squeezes each fish, observing the rate at which his fingermarks in the flesh disappear. The flesh consistency is then judged as: "beginning to lose elasticity, or, soft, not elastic, or, very soft and/or limp", according to the Manual.

What are the physical properties on which these subjective observations are based? Can they be measured objectively to provide parameters for understanding and reliably estimating the meaning of textural changes in fish flesh during storage, with respect to the extent of protein denaturation, and the acceptability of the fish to the consumer? This introductory study was undertaken in an effort to simulate the "finger" procedure in an objective manner, with the anticipation that the resulting data might be meaningful in the development of new rapid methods to grade fish quality, methods based on instrumental measurements of the physical properties of fish flesh.

The primary objectives were: 1) to establish a routine for the precise measurements of fish flesh responses to "finger-type" pressures, and: 2) to generate some actual data on the relative responses of various fish species, as well as some information on the extent of variation in response within species. (Seasonal variation in response, while considered, fell outside the scope of this study.) Desirable outcomes of this initial study would be data enabling researchers to put precise and objective time constraints on the storage life of fish under laboratory cond-

itions, thus assuring that research operations on fish flesh are carried out on quality fish; in addition, the data will evaluate deformation studies as critical indicators of the quality of fresh fish.

## MATERIALS AND METHODS

INSTRON TESTS. The initial testing procedure was devised with the use of an Instron Universal Testing Machine, equipped with a 1 cm ball probe which moved at a rate of 2 cm/min. The probe began its cycle at 0.1 cm above the sample, contacted the surface, and pushed 0.5 cm into the flesh; the direction of the probe was then automatically reversed, retracted to its starting point, and then driven downward to the same depth, and again retracted. The procedure is shown schematically in Figure 1. Calculations from the graphs provided date on the peak force exerted by the probe, the relationship between the initial peak force ($f_1$) and the second peak force ($f_2$), and the recovery of the specimen between cycle 1 and cycle 2 ($\Delta H_o$).

This procedure was followed for the preliminary test results as shown in Figure 2. However, examination of the preliminary test results revealed procedural difficulties due to ignoring the initial thickness of the sample, thus causing a much greater deformation of thinner samples. For greater precision in data analysis, a computer-operated program, developed for the Instron Testing Machine, was adopted as follows: using the same probe as described above, the computer-operated Instron moved the probe downward at 2 cm/min. to contact the specimen; upon reaching a force reading of 20g, the computer measured the thickness of the sample at that point. The probe was then moved into the flesh a distance of 10% of themeasured thickness. When this 10% deformation point was reached, the probe was withdrawn, at the same rate, until the force reading dropped to 20g (the initial preload), and the thickness again measured. The probe was reversed and again forced into the sample to a new 10% deformation and withdrawn. This entire procedure is shown schematically in Figure 3.

The information provided by the computer from this procedure was as follows: initial thickness of the sample, thickness at the beginning of the second cycle, peak force for each cycle, the energy input for each cycle, the energy returned for each cycle, and the hysteresis ($\Delta H_o$), or loss factor for each cycle. A sample printout from the computer-aided procedure is shown in Figures 4 and 5.

At this point it should be noted that the extent of deformation and the magnitude of the forces involved was chosen not only to be a non-destructive test, but also to conform as nearly as possible to the actual deformation and forces in the "thumb" testing procedure of a fish inspector.

pH MEASUREMENTS. In order to obtain some biochemical evidence correlating internal changes with the texture of the specimens, the initial pH of the fish was determined with an Orion digital

read-out pH meter, using a surface electrode. During the course of the study,a penetration probe was also introduced; in addition, pH changes during the experimental storage time were recorded.

TEST SPECIMENS. Preliminary tests were conducted on cod, bluefish, flounder, and hake fillets, obtained as fresh as possible from local sources (usually in the laboratory on the day after the boats landed). Samples were measured, weighed and pH tested before iced storage. In the later studies, whole cod and flounder were used; the cod were cut, leaving a body length of about 25 cm. Between tests the fish were stored on ice in closed containers at approximately $1^{\circ}$ C.

## RESULTS AND DISCUSSION

In the preliminary studies undertaken in March, 1982, we obtained fresh cod, bluefish, flounder and hake. Using the Instron (manually operated) we conducted deformation studies on these filleted specimens over 10 days of storage on ice at $1^{\circ}$ C. The results of this run are shown in Figure 2, where the $\Delta H^{\circ}$ values (recovery between cycles) are compared. (In evaluating these results, it is important to consider that the larger the value of $\Delta H^{\circ}$, the less recovery; the smaller values of $\Delta H^{\circ}$ indicate greater recovery between cycles, hence greater resilience or elasticity).

This tesing procedure revealed a general trend in textural change over storage time common to the four specimens tested; $\Delta H^{\circ}$ values were higher at the start, dropped to a low point 4-6 days after storage began, and then rose somewhat again, till the end of the run. While the codfish tested showed considerable variation in recovery, its values for $\Delta H^{\circ}$ over the first 6 days were close to those of hake and bluefish. However, the consistently higher values for flounder indicated less recovery of its flesh between test cycles, in comparison with the other species. In evaluating these data it should be noted that during the initial Instron testing the probe was forced a standard 0.5 cm into the specimen; in the case of a relatively flat thin fish as flounder, this 0.5 cm represented a much greater percentage of the total thickness of the specimen than for the other species tested.

In order to obtain readings more consistent with the thickness of the specimen, and to escape the difficulties of reading the values from the graphs, the remainder of the study was conducted using the computer-aided Instron procedure. Figure 6 exemplifies some of the results obtained over the spring and summer; as shown, the relative recovery of the flounder now appears greater than that of cod. In April the recovery values for cod were comparable to those obtained in March, $\Delta H^{\circ}$ values between 0.18 and 0.13 for the 10 day period. However, by August, the general range of $\Delta H^{\circ}$ values for cod were higher, between 0.19 and 0.26 (probably a seasonal influence ( 2)).

The computer-aided operation also generated data on the maximum stress and energy values for both cycles (Figures 4 and

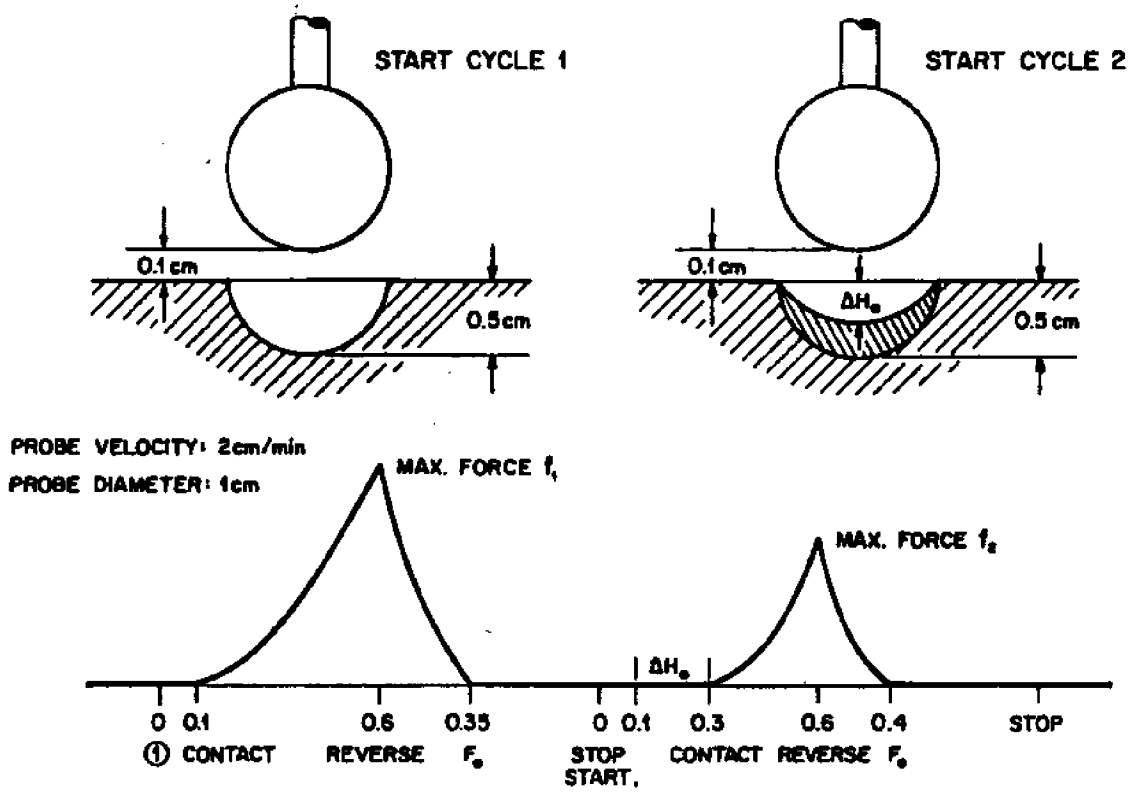
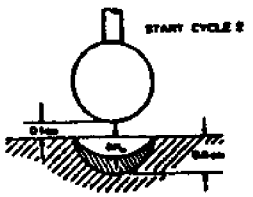



Figure 1. Initial Instron testing procedure



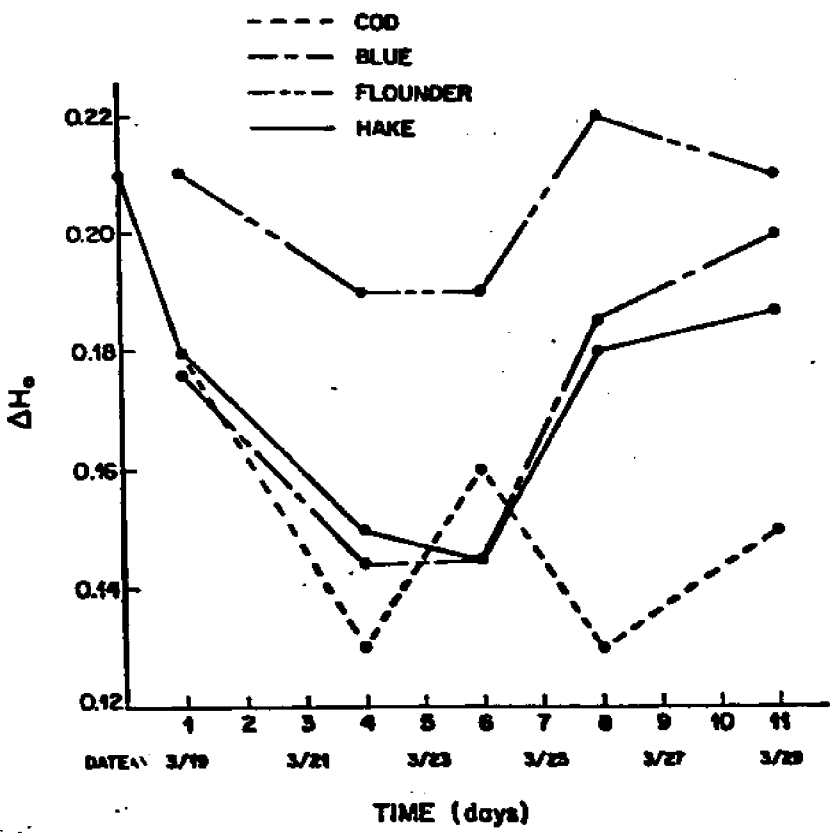



Figure 2. Changes in $\Delta H_o$ values during iced storage for cod, hake, bluefish, and flounder.

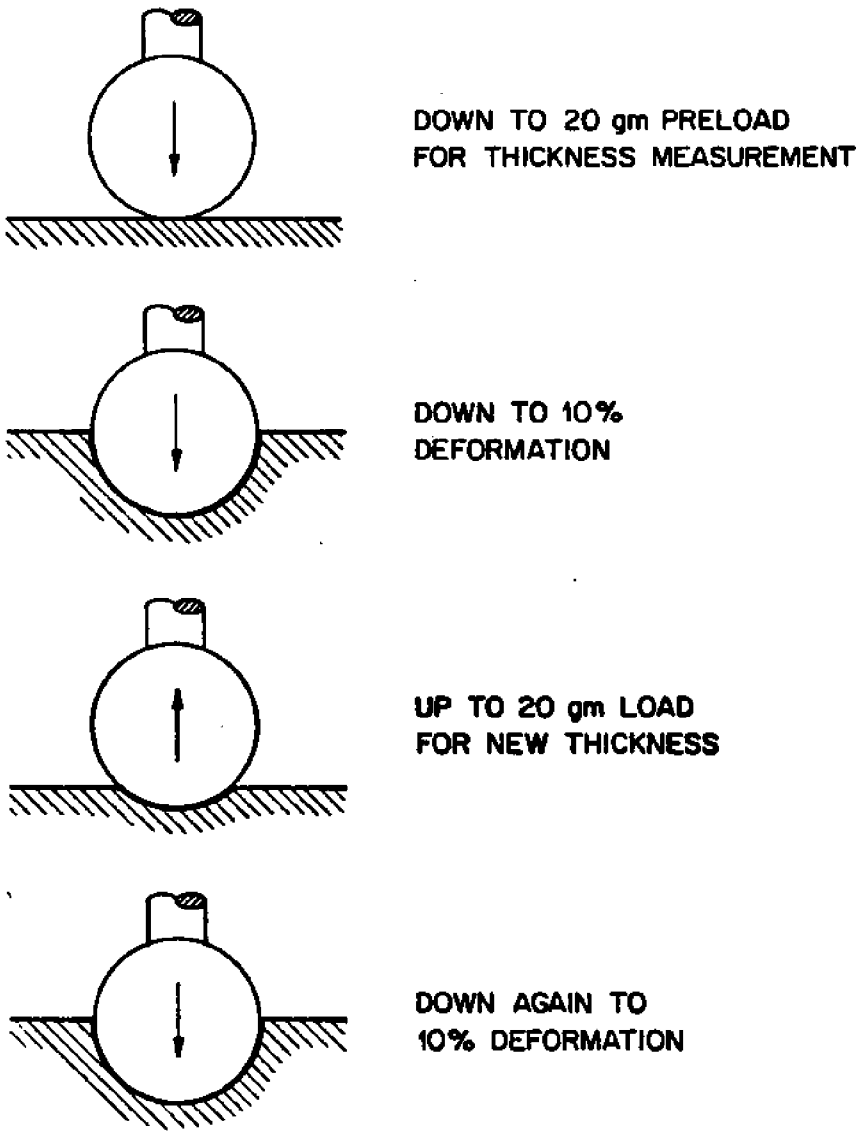



Figure 3. Selected Instron testing procedure

(Computer controlled)

C.S.Dim. : A1

Sample ID : COR 41

F.S.L. : 0.5
G.L. : 6.0

Full Scale Load : 0.50

| | | Cycle Number | Initial Thickness | Stress at Max. Strain |
|---|---|---|---|---|
| 0.076 | 0.100 | 1 | 5.716 | 2.377 |
| 0.090 | 0.137 | 2 | 5.471 | 4.041 |
| | | | | Cohesiveness : |
| 0.064 | 0.091 | 1 | 5.559 | 1.934 |
| 0.122 | 0.172 | 2 | 5.317 | 4.025 |
| | | | | Cohesiveness : |
| 0.044 | 0.066 | 1 | 5.170 | 2.059 |
| 0.115 | 0.166 | 2 | 4.941 | 4.075 |
| | | | | Cohesiveness : |
| 0.047 | 0.068 | 1 | 4.871 | 2.031 |
| 0.111 | 0.152 | 2 | 4.682 | 3.712 |
| | | | | Cohesiveness : |
| 0.036 | 0.056 | 1 | 4.894 | 1.535 |
| 0.098 | 0.132 | 2 | 4.689 | 2.855 |
| | | | | Cohesiveness : |

Cycle 1

| | | |
|---|---|---|
| Mean | 5.242 | 1.991 |
| Low | 4.871 | 1.535 |
| High | 5.716 | 2.377 |
| Variance | 0.147 | 0.087 |
| Std.Dev. | 0.384 | 0.295 |

Figure 4. Sample computer printout of Instron testing procedure

Cross Sectional Area : 1.00

Gauge Length : 4.00

| Apparent Modulus | Strain Energy Loading | Strain Energy Unloading | Poissons Ratio | Hysteresis Loss |
|---|---|---|---|---|
| 21.995 | 0.094 | 0.043 | 0.000 | 0.550 |
| 39.405 | 0.211 | 0.109 | 0.000 | 0.486 |
| 2.236 | Relaxation Index : | 2.236 | | |
| 17.989 | 0.078 | 0.034 | 0.000 | 0.560 |
| 43.138 | 0.184 | 0.093 | 0.000 | 0.494 |
| 2.359 | Relaxation Index : | 2.359 | | |
| 15.473 | 0.089 | 0.038 | 0.000 | 0.573 |
| 41.092 | 0.184 | 0.094 | 0.000 | 0.478 |
| 2.070 | Relaxation Index : | 2.070 | | |
| 15.796 | 0.084 | 0.040 | 0.000 | 0.522 |
| 37.110 | 0.170 | 0.092 | 0.000 | 0.461 |
| 2.039 | Relaxation Index : | 2.039 | | |
| 11.866 | 0.067 | 0.031 | 0.000 | 0.537 |
| 30.371 | 0.139 | 0.075 | 0.000 | 0.459 |
| 2.055 | Relaxation Index : | 2.055 | | |
| 16.624 | 0.083 | 0.037 | 0.000 | 0.548 |
| 11.866 | 0.067 | 0.031 | 0.000 | 0.522 |
| 21.995 | 0.094 | 0.043 | 0.000 | 0.573 |
| 13.838 | 0.000 | 0.000 | 0.000 | 0.000 |
| 1.720 | 0.010 | 0.004 | 0.000 | 0.020 |

Figure 5. Sample printout of Instron testing procedure.

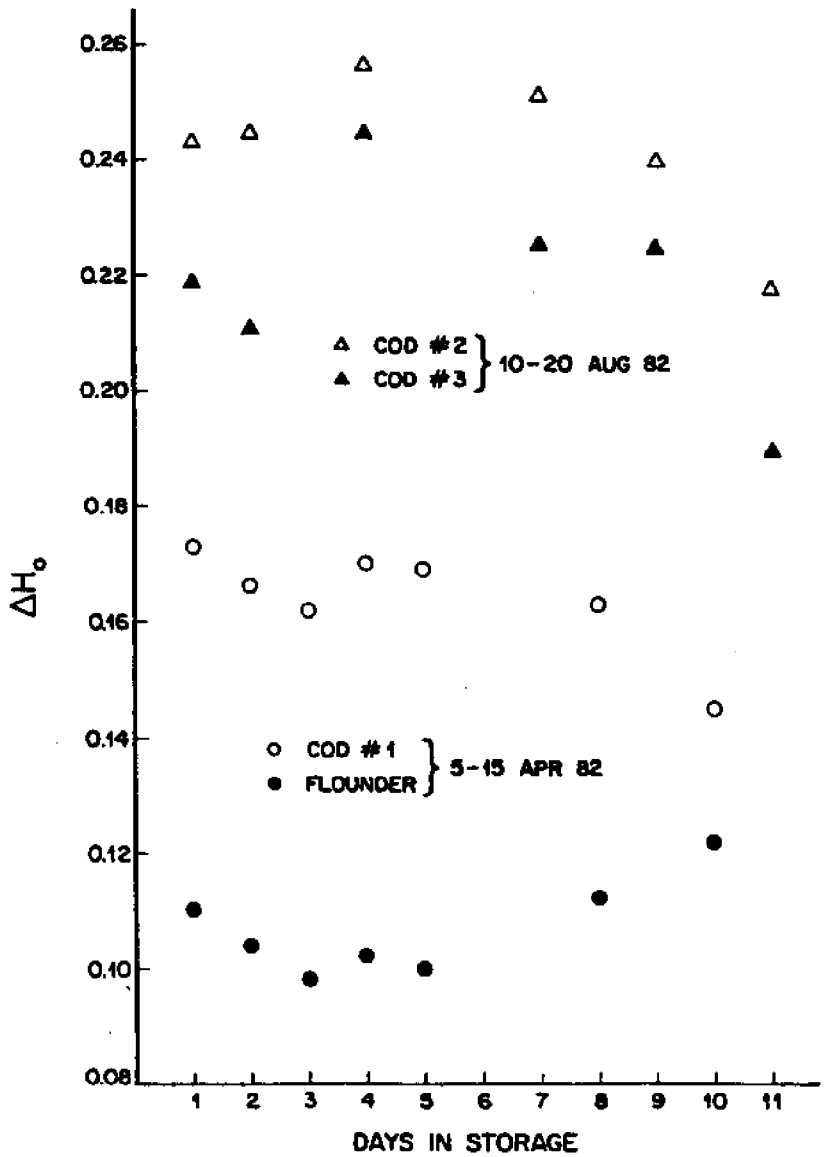



Figure 6. Changes in $\Delta H_o$ values during iced storage. (Computer-aided Instron).

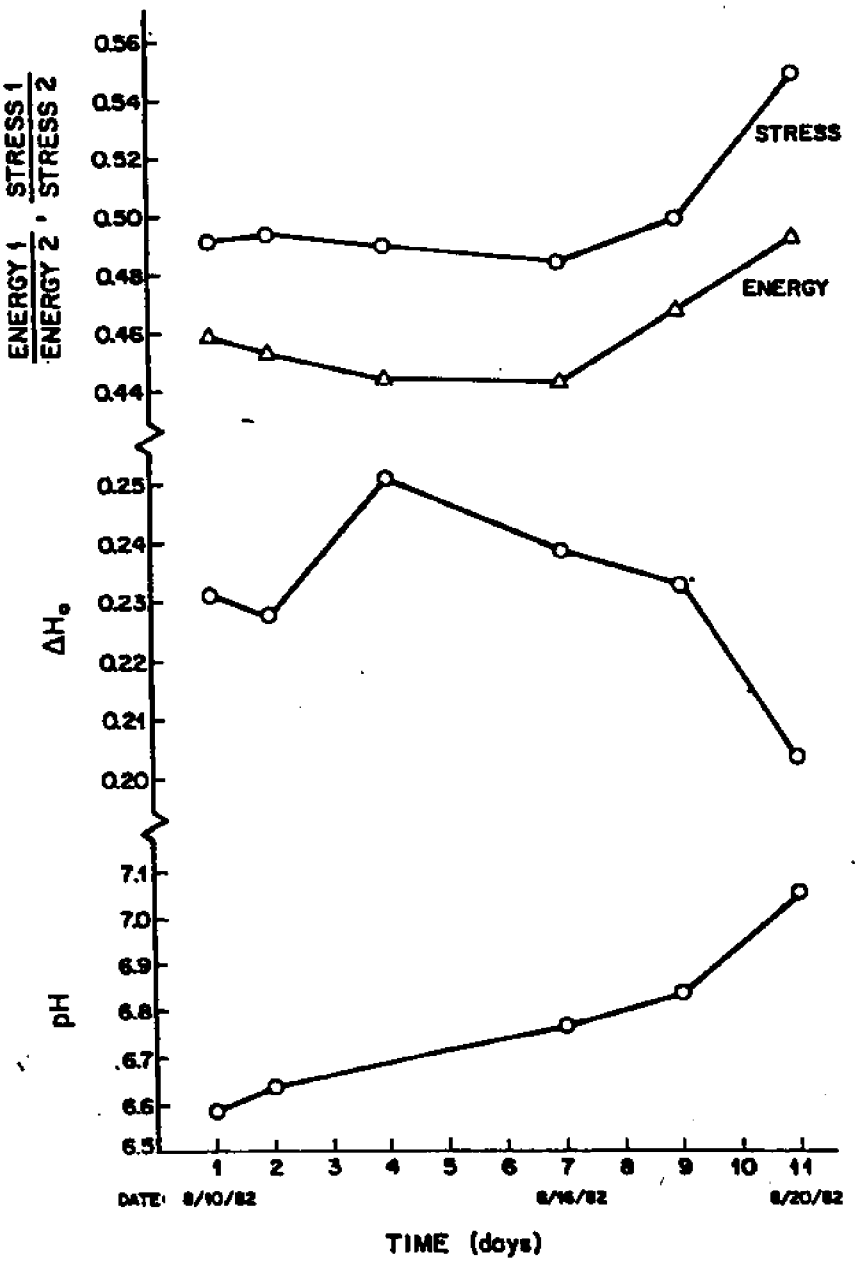



Figure 7. Changes in stress and energy relationships, $\Delta H_o$ and pH values in cod during iced storage.

5); the relationship between cycle 1 and cycle 2 values were calculated, and the change over storage time plotted (Figure 7). Over this 10 day period in August, the stress and energy curves (obtained here from cod) parallel each other closely, with low values over the first few days, gradually increasing after 7-8 days. The $\Delta H_o$ values tend to diminish, a trend also observed in values obtained for cod in April. pH values over this storage period increased steadily.

With the objective of establishing a range of values for fish texture that might be encountered by an inspector during summer operations. the next storage experiment was extended to 17 days (Figure 8, 10). The relationship between cycle 1 and cycle 2 with respect to energy showed a peak at about the 10th day of storage, much as seen in the earlier run (Figure 7). However, extending the storage time revealed that after the 10th day, these values fell off rapidly. The range in absolute values between the 2 experiments in $\Delta H_o$ values was quite close, between 0.40 and 0.49. The stress values follow the same pattern as the energy values. During this 17 day run, we again observed a clear trend toward increasing pH values (Figure 9). The relatively steep drop in $\Delta H_o$ values between the 2nd and 5th days corresponds to the findings of Hennings (1); Hennings used as electronic device to measure specific resistance of muscle tissue of cod and found greatest changes during the first 4 days of storage.

As mentioned previously, larger values of $\Delta H_o$ indicate greater recovery or resiliency. However, the findings of this study from objective measurements by computer-aided Instron indicated that in stored cod, resiliency increased over a 10-day period, and then began to decrease. The pH data offered some biochemical correlation with the storage period, increasing sharply after about 2 weeks storage when serious spoilage began. We conclude that simple compression testing does not clearly correspond with the freshness of fish; however, these results do raise the question of what physical attribute is evaluated by the "finger" test, and point up the need for further work on the variation in elasticity within species and the range in variation in responses between species. Textural changes in raw fish flesh during storage may be measurable by a more dynamic test now under consideration which involves the internal flesh and is less affected by the skin.

## CONCLUSION AND REFERENCES

The computer-aided Instron testing procedure was found necessary to perform consistent, precise, deformation tests on fish flesh. Otherwise variations in the thickness of the specimens can influence the degree of recovery.

Parameters that indicated changing trends during storage were: stress 1/stress 2, energy 1/energy 2, and $\Delta H_o$. The trend indicated by the recovery values for cod in this study suggests that in fact resilience may increase during storage.

Differences in responses between fish and differences in response apparently related to seasonal factors were noted.

REFERENCES


1. Henning, C. A rapid new electronic process for determining the freshness of salt-water fish. Zeitchrift fur Lebensmittel-Untersuchung und Forscung, 119 (6): 25 Apr. 1963.

2. Love, R.M. Variability in Atlantic Cod (Gadus morhua) from the Northeast Atlantic; a Review of Seasonal and Environmental Influences of Various Attributes of the Flesh. Journal of the Fisheries Research Board of Canada, 32 (12): 2333-2342. 1975.

3. Witfogel, H. in Sanitary Regulations for Fish and Fish Products, OECD Document # 51, pp. 166-167.

4. NMFS Fishery Products Inspection Manual, Pt. II, Ch. 18, Sec. 01, pp. 1-22. 1975

5. Proceedings of the Seafood Science and Technology Workshop, 1982, University of Delaware, Sea Grant College Program, Newark, Delaware. 19711. 1982.


END

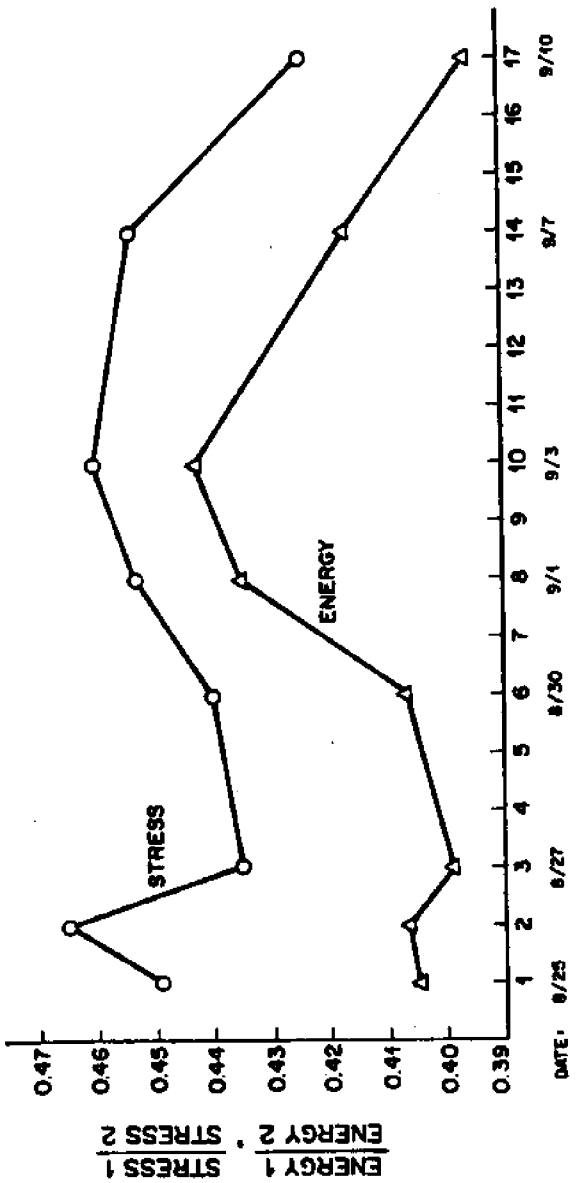



Figure 8. Changes in stress and energy relationships in cod over storage time.

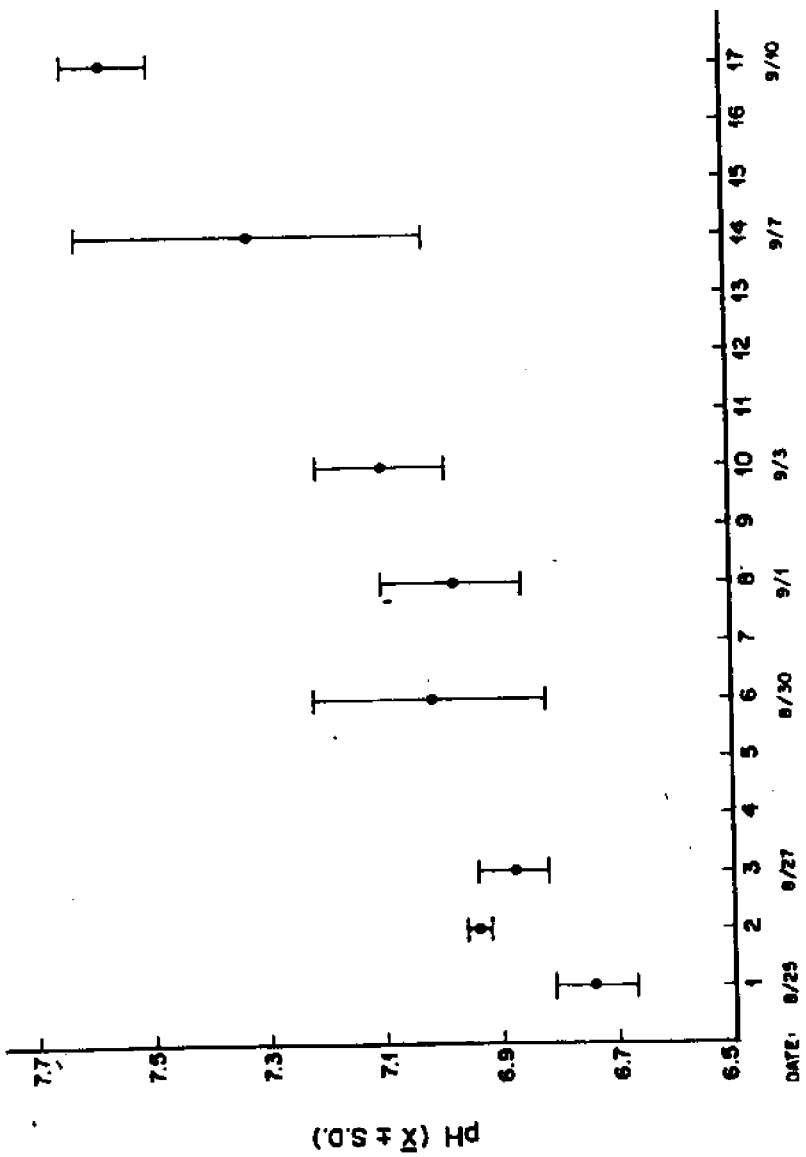



Figure 9. Changes in pH values over storage time in cod.

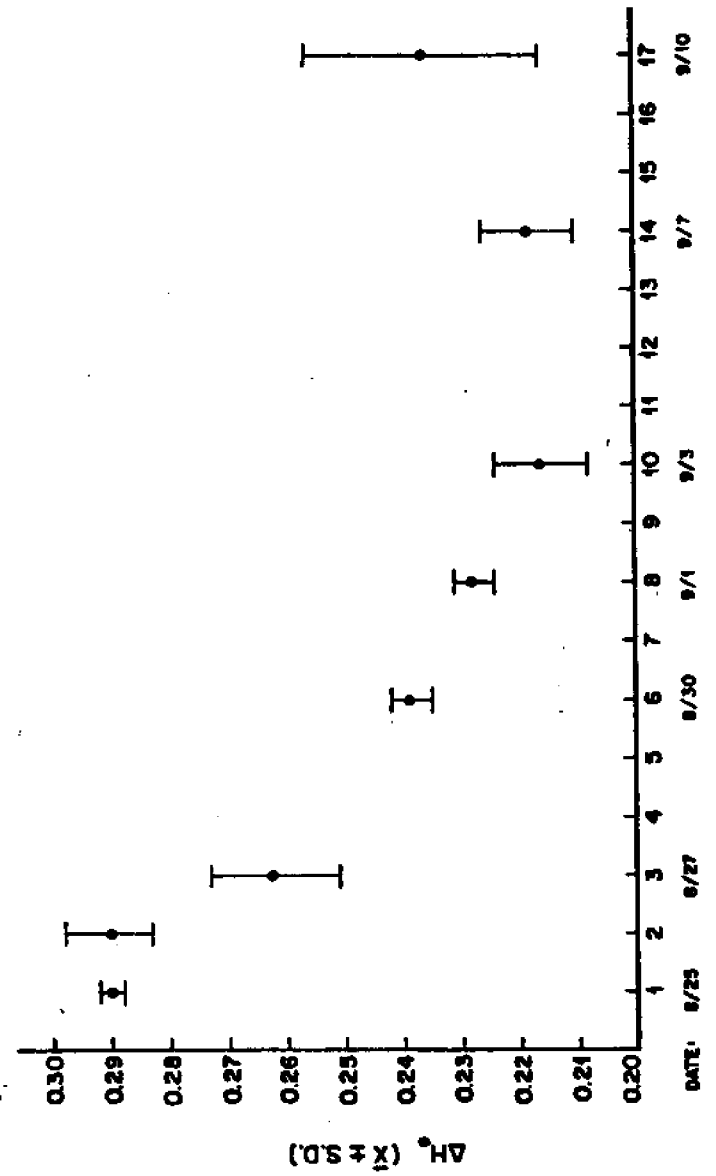



Figure 10. Changes in $\Delta H_o$ values during storage in cod. (Computer-operated Instron.)

# TEXTURAL VARIATION WITHIN COOKED FISH FILLETS

Ronald A. Segars[1], Ernest A. Johnson[2]

[1]Research Physicist
U. S. Army Natick Research & Development Lab
Natick, MA

[2]Associate Professor
Food Engineering Department
University of Massachusetts
Amherst, MA

INTRODUCTION. The instrumental evaluation of the textural properties of fish flesh has received very little attention in the past, due in part to the difficulty of making instrumental texture measurements on anisotropic products of complex structure such as that of fish flesh and in part because fresh fish flesh, in contrast to beef muscle, seldom becomes too tough to masticate readily. However, objective instrumental measurements of the textural properties of fish flesh can be of tremendous value in many studies for they provide a basis for correlation with sensory judgments, and thus ultimately, for prediction of consumer perception of texture. Using this data, underutilized species can be grouped along with more commonly known species according to their prospective processing and consumer edibility characteristics. Furthermore, a data bank of species specific textural properties provides a means to evaluate the effect of different methods of catch, processing, storage, and related conditions on consumer acceptance of the textural properties of the final product.

Stringent sampling methods are required in making instrumental measurements of textural variation in fish flesh. Freshness is essential in view of the highly perishable nature of fish flesh. Effects of natural variations due to size, season, feeding habits and other environmental factors must also be minimized. Both the instrument and the accompanying sensory panel must evaluate similar test samples; since both methods are sample destructive, large areas of flesh of uniform texture are necessary to minimize variations between instrument and panel findings. Clearly, non-destructive rheological test procedures and use of very small specimens for pH and chemical analyses would improve agreement among the various test procedures (sensory vs instrumental) by reducing the experimental error due to variation between samples. However, these methods are not yet available. Consequently, more information on the range of textural variation within the test species and within individual fish to identify regions of uniform texture is a prerequisite to obtaining textural evaluations of greater correlation and predictive capabilities.

## METHODS AND MATERIALS

### Sample Preparation

Fresh, whole codfish were purchased from a local dealer (Capt'n Bob's, Route 126, Framingham, MA) on the day that he received the fish, which in most cases was the day the fish were landed. Fish were examined carefully to select desired sizes (between 2 kg and 5 kg) and to avoid any apparent defects or wounds. For some studies pH measurements were made at the dealers, to meet particular experimental requirements (low pH, for example).

At the testing laboratory each fish was weighed and its overall length measured before being hand filleted and skinned. The pH was measured in three locations on each fillet using an Orion pH meter with either a surface, needle penetration, or immersion probe. When using the immersion probe, 10 g of flesh were well mixed in 10 ml of water to make a slurry; no preparation was required for the other probes.

### Cooking

Fish samples were cooked in plastic bags in a 70°C water bath. A thermocouple monitored internal temperature of the fillet; at 69°C the sample was removed from the water bath. In most cases the whole fillet was cooked, except for a portion toward the tail end where flakes were too small for the instrumental testing. In a few tests, the fillet was cut into portions about 10 cm long and each portion was cooked in a separate bag.

The bags were laid flat on a horizontal rack and covered with a flat perforated plate of sufficient weight to keep the fish submerged. The bags were not sealed but their tops were held above the water line and clamped to the side of the cooking vessel.

### Test Specimens

Test specimens were identified by first separating the fillet or portions into three longitudinal sections--dorsal, central, and ventral, as shown in Figure 1. The central portion was further subdivided into an upper and lower segment. Individual flakes of each segment were then gently separated by knife point, with care to maintain the order of the flakes within each longitudinal segment. Each flake of each segment then served as a single test specimen. Love (2) has shown for cod, that structure remains unchanged (number of flakes and number of fibers is constant) with age and size, and therefore the structural pattern shown in Fig. 1 applies to all cod. It is expected that similar characteristic patterns exist in other species.

### Instrumental Evaluation

A computer controlled Instron Universal Testing Instrument equipped with a punch and die test cell (4) was used for all texture measurements. A 1 cm diameter flat end punch was driven through the test

SCHEMATIC DIAGRAM OF A COD FILLET

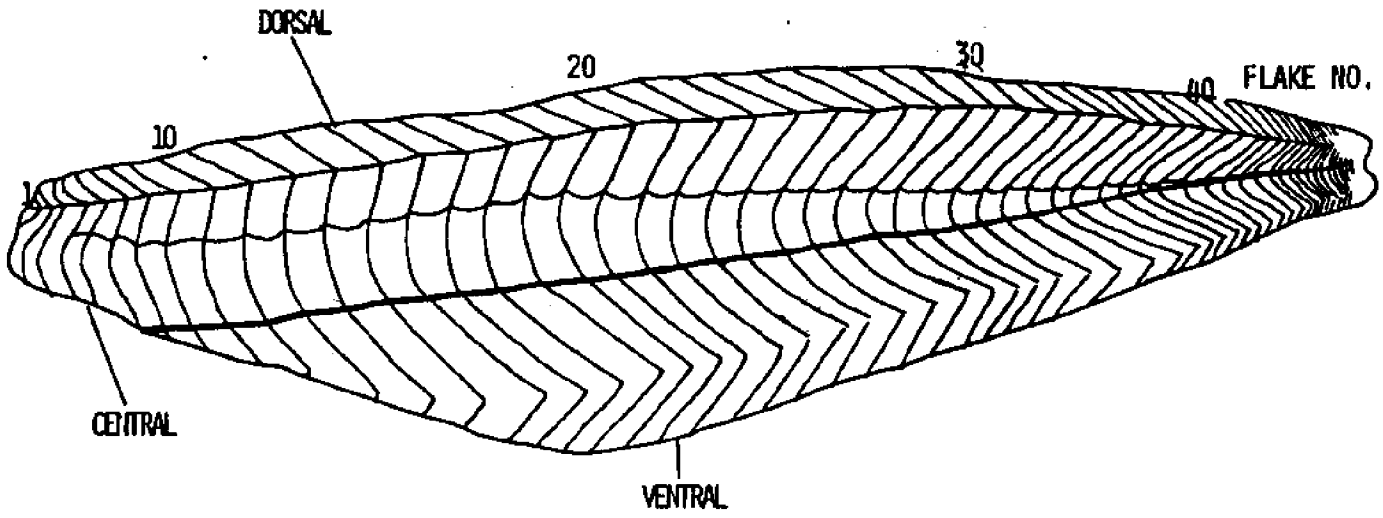



Figure 1. Schematic diagram of a cod fillet.

specimen and into the die (hole in the support plate) at a constant rate of 2 cm/min. The computer program determined the thickness ($h_o$), maximum shear stress $\left( \gamma_m = \frac{Pm}{\pi d h_o} \right)$ stiffness $\left( S = \frac{m}{\pi d} \right)$ and strain at peak force $\left( \varepsilon_m = \frac{Dm}{h_o} \right)$ from the force-deformation characteristics of the specimens. A 20 g preload was considered necessary to provide good contact between flakes, the plate, and the punch, and to assure accurate and reproducible measurements of sample thickness ($h_o$). The computer used punch location at the time the force first reached 20 g to determine $h_o$. The longest linear region between 5 and 35 percent of the maximum force (Pm) was taken as the slope (m) for the determination of S. The punch diameter is d and Dm is the sample deformation at the time Pm is reached (Figure 2). Each specimen described above was evaluated following this procedure. This provided between 40 and 90 measurements on each fillet.

## RESULTS AND DISCUSSION

Figures 3(a)(high pH), 3(b)(intermediate pH), and 3(c)(low pH) show the maximum shear stress for each flake plotted vs the relative position of the flake (the location of the first flake evaluated, proceeding from head to tail, is assigned 0 and the last flake evaluated is assigned 1.0. All intermediate flakes are placed at equal intervals in between). These plots show clearly the wide range of textural properties that may exist within a single fillet, particularly those of low pH.

With this two-fold or larger spread in values, a population of randomized samples would generally exhibit a large standard deviation. When studying the effect of a storage treatment, process variable, or seeking sensory-instrumental correlations, a large standard deviation within the treatment groups greatly reduces the possibilities of success and at best requires a larger number of measurements to be performed to obtain statistically significant results. Obviously, if regions of uniform textural properties can be defined, such studies could be conducted with much less effort and with greater probability of success. Also, efforts to group fish species according to their edibility characteristics might benefit from data on textural variations. If large regions of uniform texture exist, it might be better to let this region represent the textural properties of the species rather than to alter the mean value for this large region, which may be represented by relatively few measurements, e.g. a region of large flakes, by including data on small flakes of greatly different texture.

The graphs of Figure 3 show trends that appeared in a number of fillets not included here. In all cases the dorsal section gave the highest shear stress values (toughest region) with the difference increasing as the pH went lower. This trend of increasing variability with lower pH was observed in all data. In many cases the ventral section was tougher than the central section but differences were small and were not observed

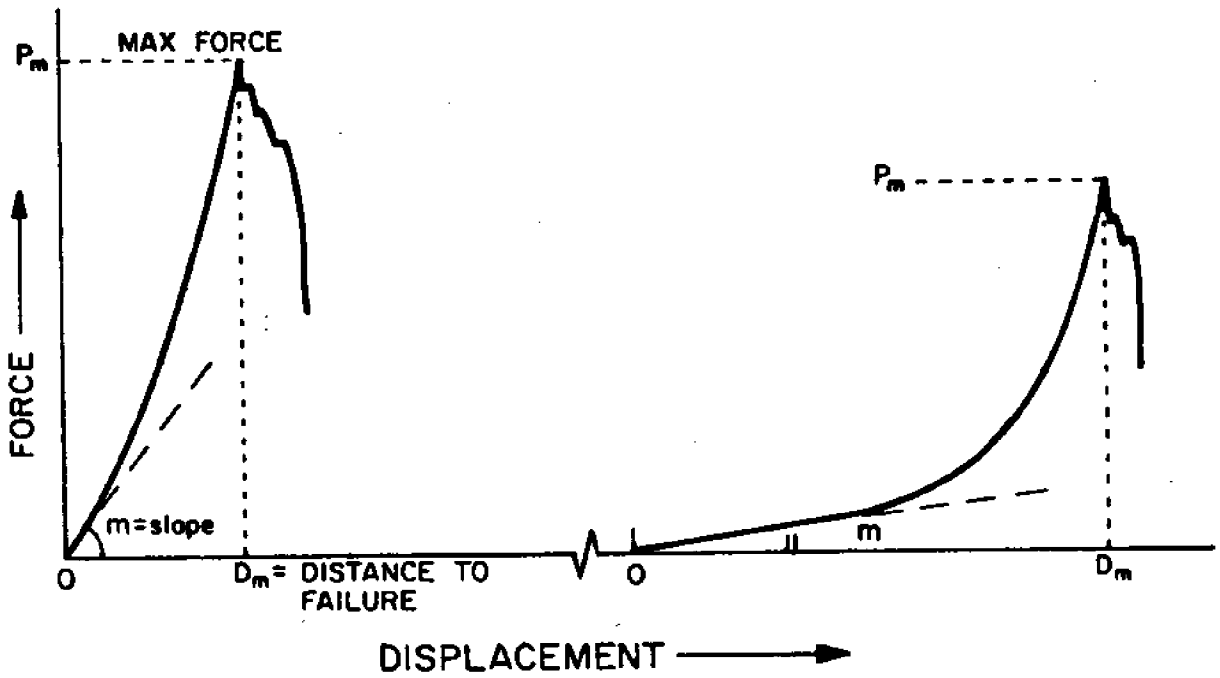



Figure 2. Typical force-displacement curves produced by the punch and die test cell on cooked fish flakes. Features used in subsequent analyses are indicated. The curve on the left represents a firm textured fish; the one on the right a soft fish.

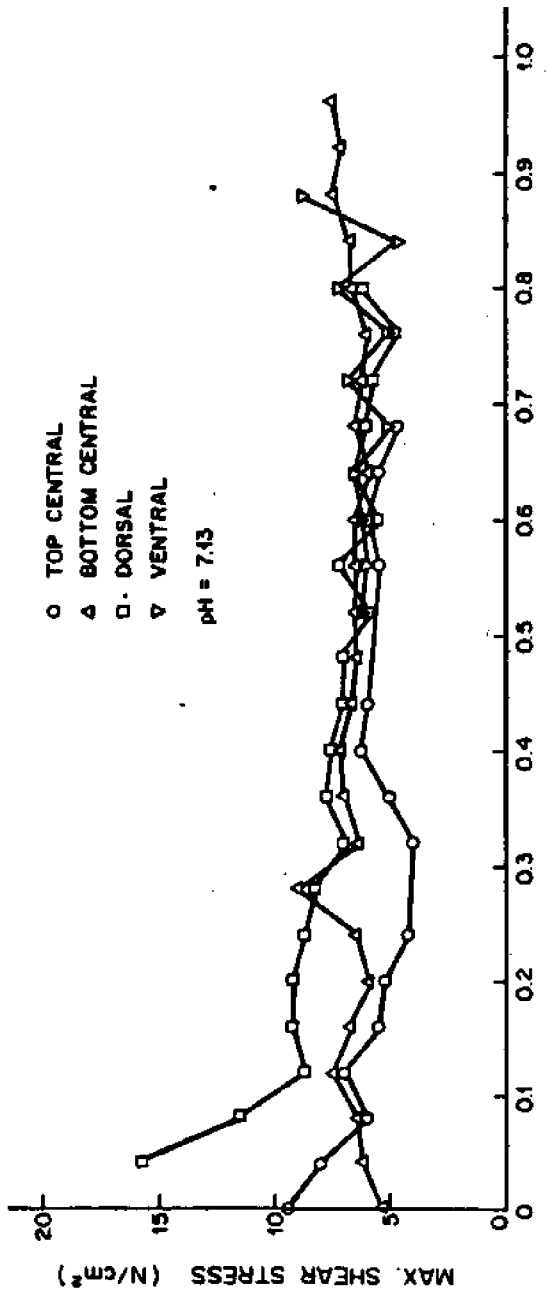



Figure 3a. Maximum shear stress for each flake plotted versus flake position for a high pH fillet.

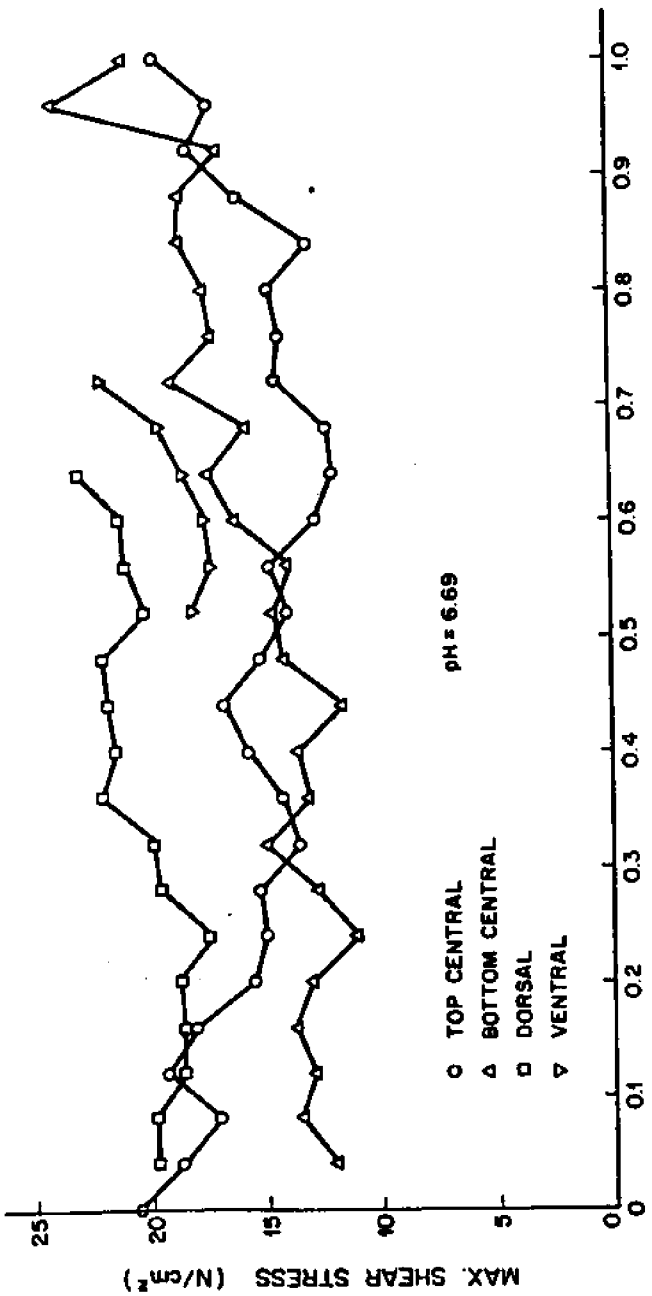



Figure 3b. Maximum shear stress for each flake plotted versus flake position for an intermediate pH fillet.

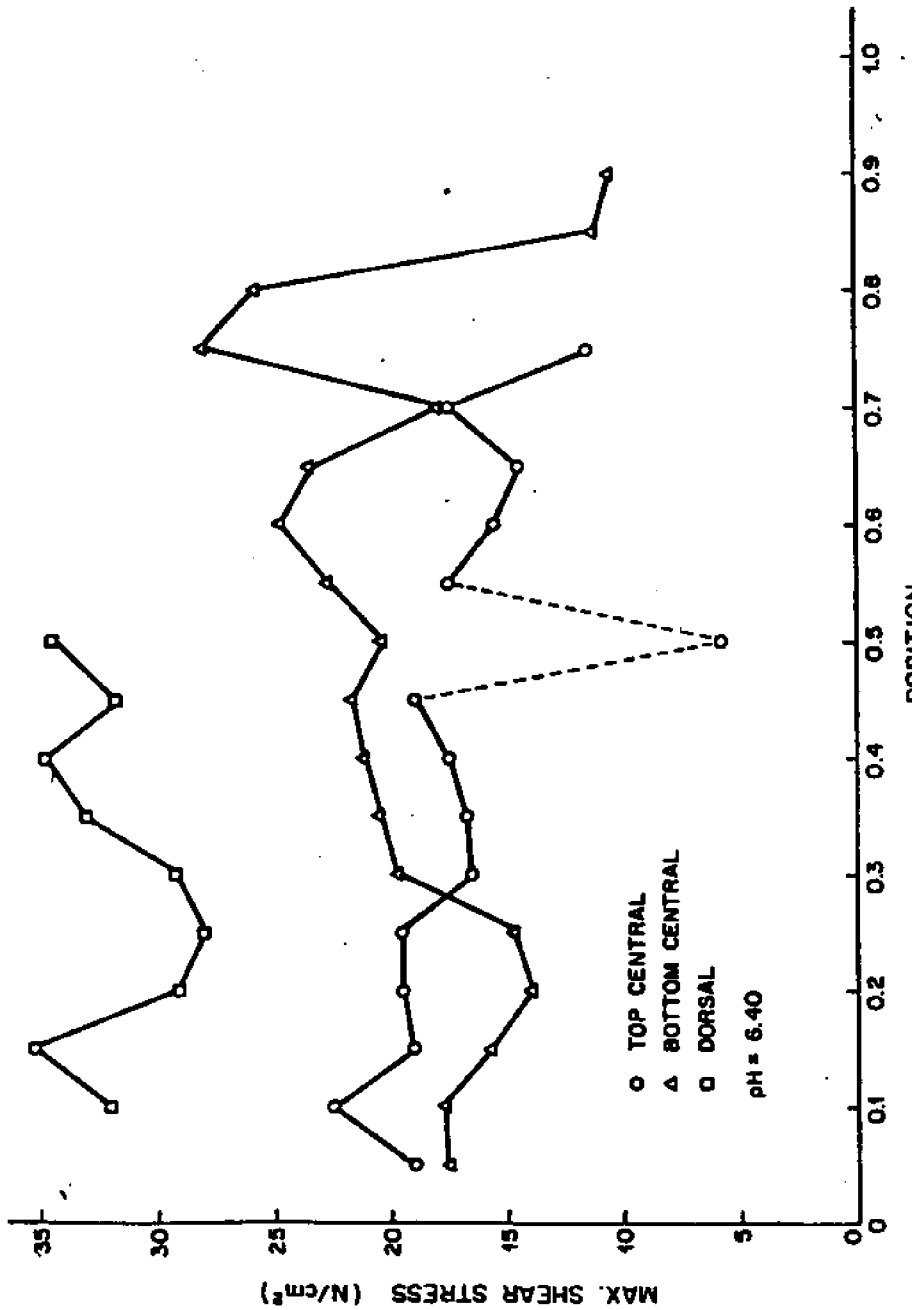



Figure 3c. Maximum shear stress for each flake plotted versus flake position for a low pH fillet.

at high pH. Usually the top central section was tougher than the bottom central near the head end of the fillet and more tender toward the tail. Also when considering all data it appears that the mid-length of the fillet is more tender than the two ends. However, this is not shown as clearly in the data of Figure 3 as in other data obtained in this study.

The graphs of Figure 3 show that uniform areas do exist but it is difficult from these plots to determine the size of these regions. The fish texture maps of Figures 4a, 4b, and 4c show the actual maximum shear stress values determined for each flake. These data are for the same fillets plotted in the graphs of Figures 3a, 3b, and 3c respectively. From Figure 4a, which is the high pH fillet, it is clear that if the first 8 to 10 flakes are discarded (along with the tail section for which no data was obtained), the remaining 60-70% of the fillet is quite uniform. Most shear stress values in this region fall within the range from 5 to 8 $N/cm^2$ with occasional 4's and 9's. It should be noted at this point that no threshold studies have been undertaken to determine the minimum instrumental change required to produce a detectable change in sensory evaluation. However, previous sensory-instrumental correlation studies using various fish species (1) produced good correlations between the sensory attributes of Hardness and Chewiness and the maximum shear stress. The slope of the regression lines of Hardness (or Chewiness) vs maximum shear stress was 0.27. Thus a change in m of 3.7 produces a change of 1 unit in the sensory attributes of Hardness and Chewiness. This 1 unit change represents the sensitivity of an individual panelist, and is usually close to a just detectable difference. The region of Figure 4a just described would then appear uniform to a panelist. The map shown in Figure 4b has a wide spread of values in correlation with the lower pH. In this case if flakes 10 through 27 of the central section, which represents perhaps 50% of the weight of the fillet, are saved for the experiment, then most values fall in the range 13 to 17 $N/cm^2$ with occasional 12's and 18's. This region would appear quite uniform to a panelist. In Figure 4c three uniform regions of approximately equal size can be seen: a) the dorsal section, b) the upper central section minus the first nine flakes, and c) the lower central section minus the first nine flakes. Within each region values are quite uniform and would appear uniform to a panelist. Between these regions however, the changes are quite large (5 or 6 units in one case and about 10 units in the other) and are probably detectable by a panel.

From the instrumental texture profiles of Figure 4 it appears that uniform regions of a reasonable size to provide test specimens for textural studies can be found in most fish fillets.

The measurement of pH may be the best non-destructive test to use in selecting those fillets most likely to have large uniform regions. Its measurement and correlation with many textural observations is important. Figure 5 plots the average pH values for each fillet vs the average shear stress values for each region (dorsal, ventral, upper and lower central). This plot shows the expected trend (3), both overall and for individual regions, of decreasing toughness (lower shear stress values) with increasing pH.

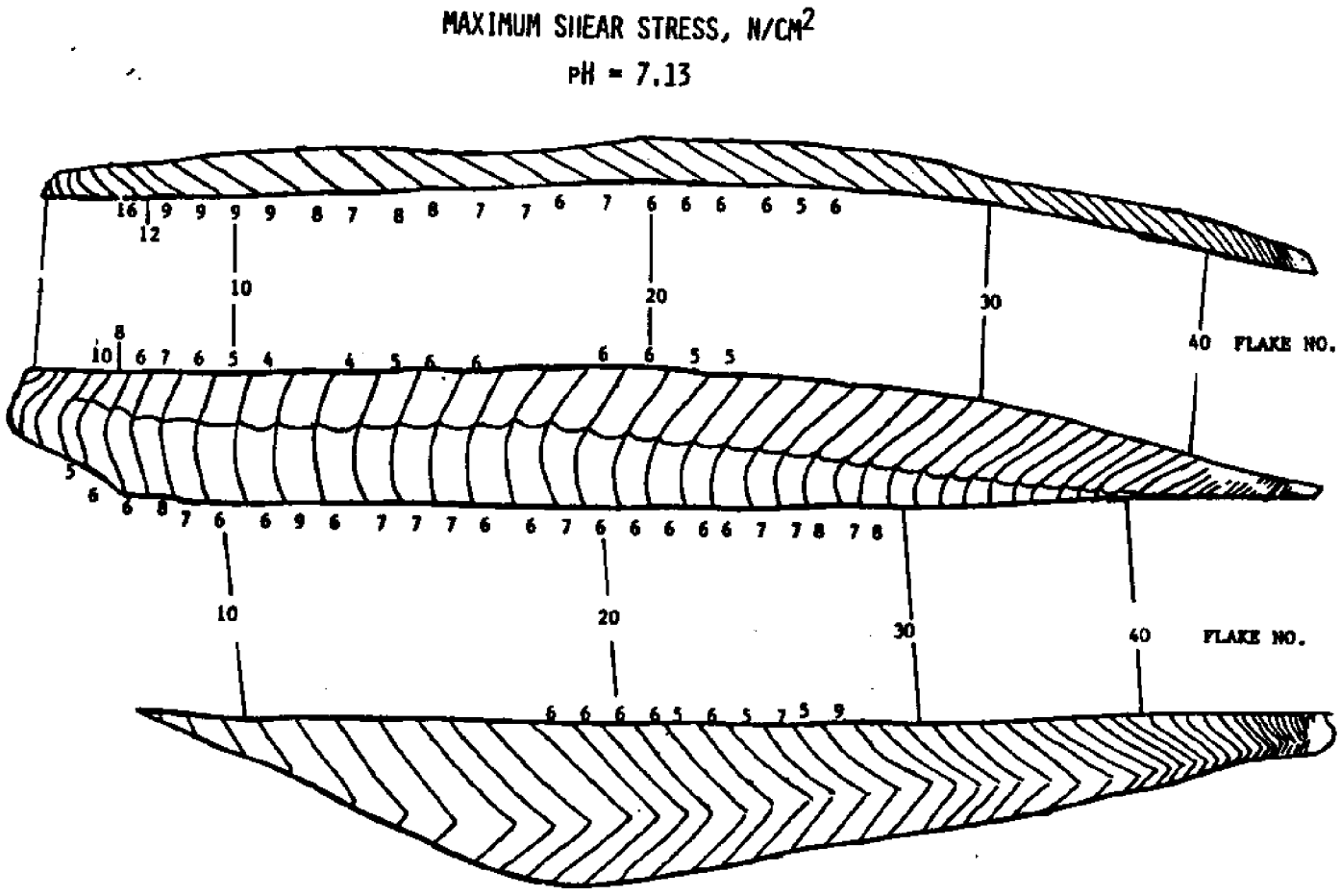



Figure 4a. Fish texture map of maximum shear stress for each flake of a high pH fillet.

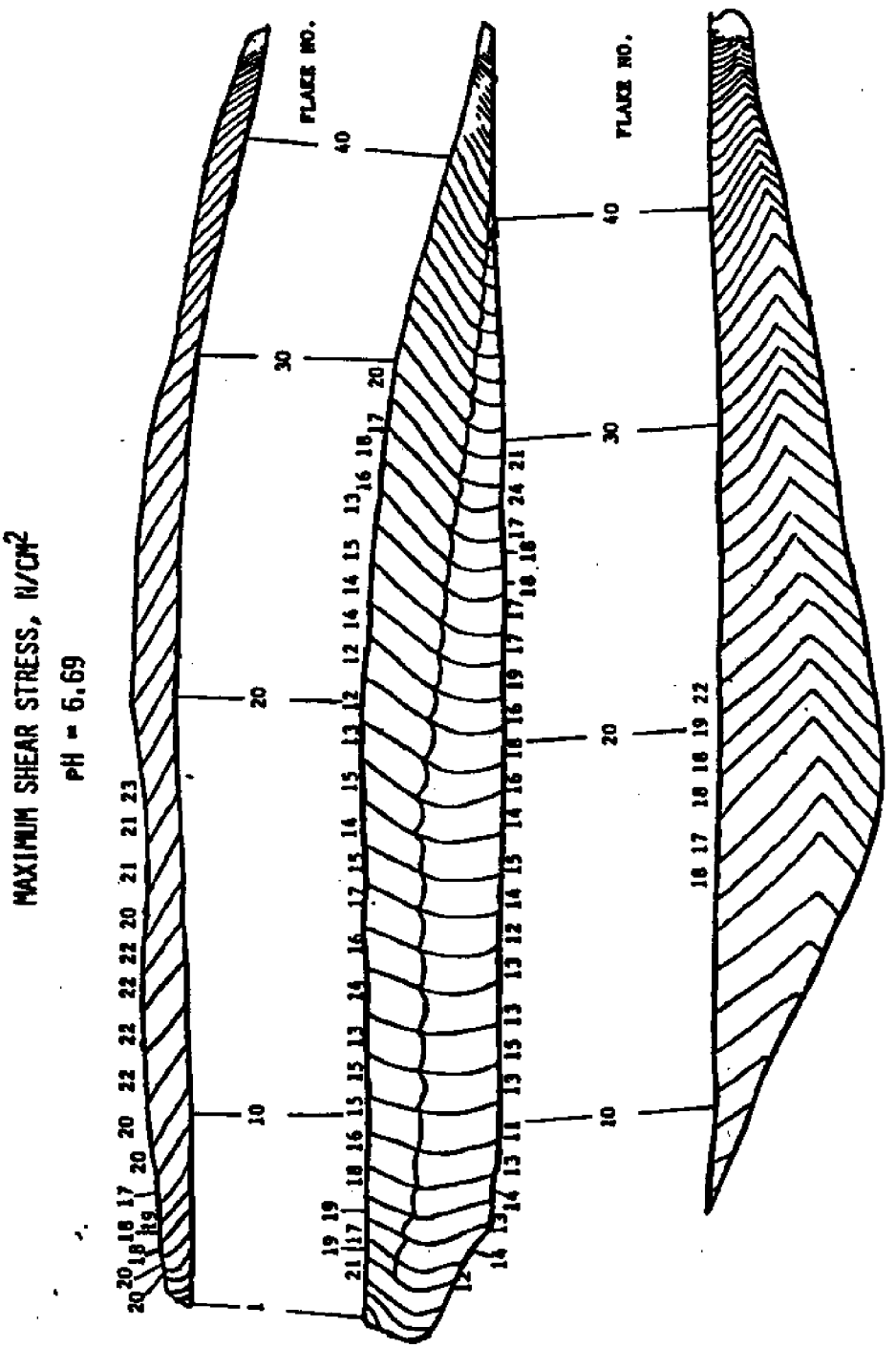



Figure 4b. Fish texture map of maximum shear stress values for each flake of an intermediate pH fillet.

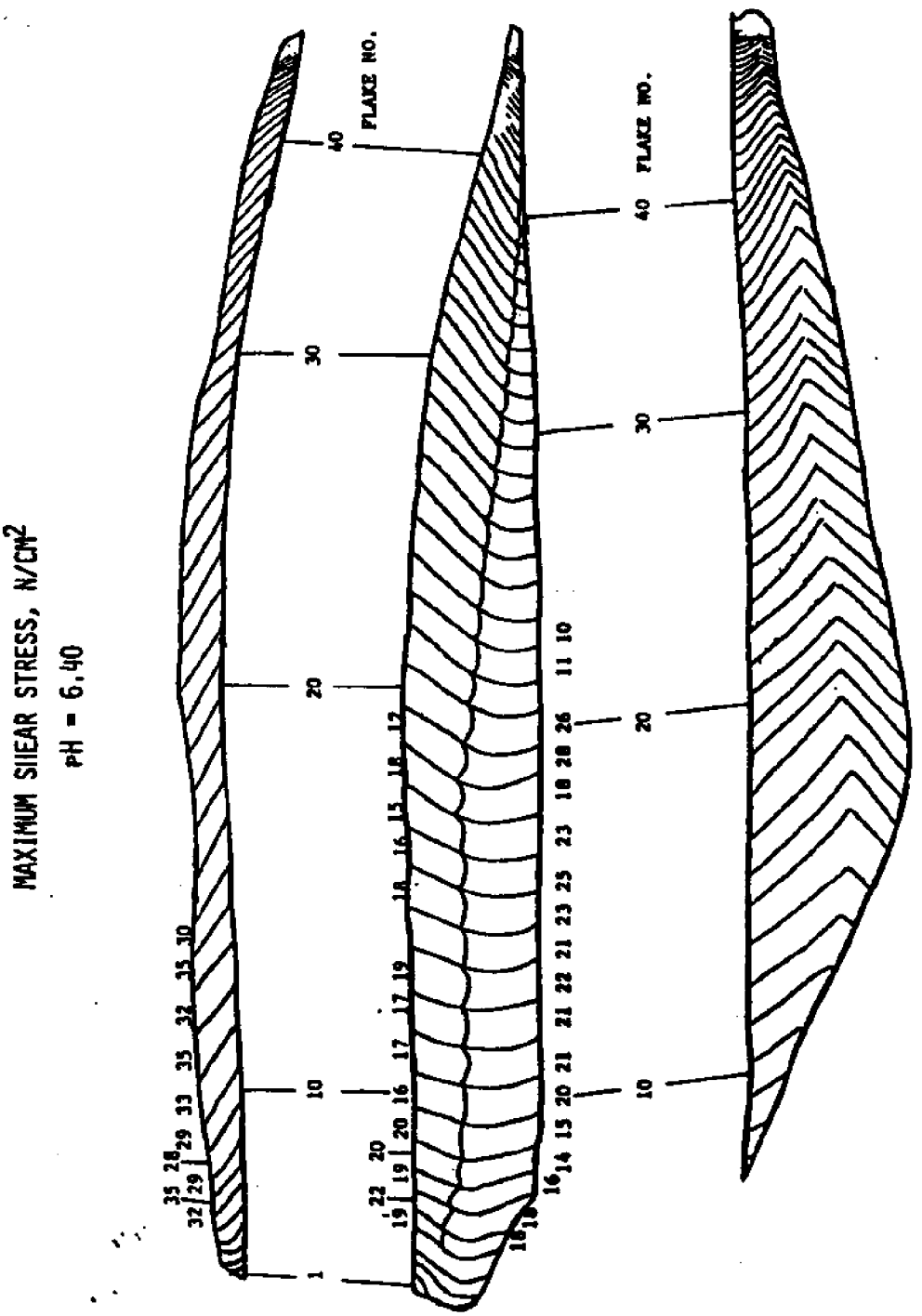



Figure 4c. Fish texture map of maximum shear stress values for each flake of a low pH fillet.

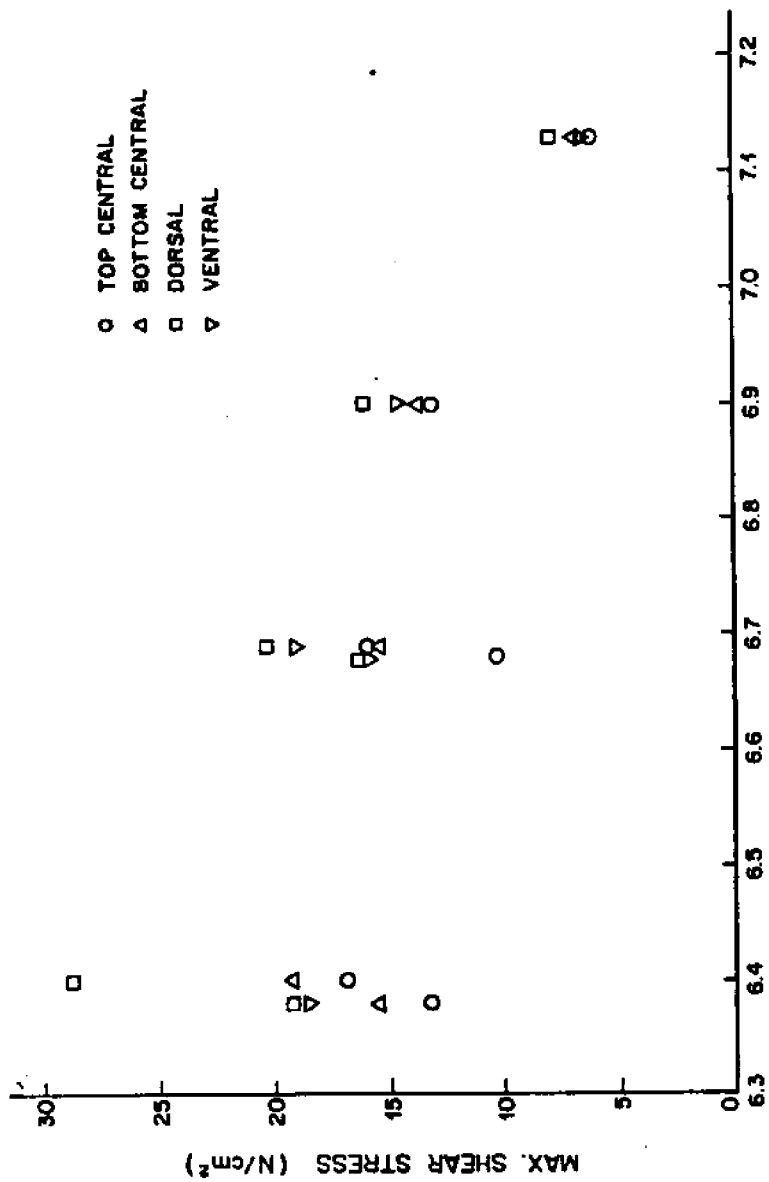



Figure 5. Average pH values for each fillet versus the average shear stress values for each region (dorsal, ventral, upper and lower central.)

Since pH correlates well with overall texture, it seemed probable that the textural variations within a fillet might also be correlated with pH changes. A low pH fillet was selected for mapping of both pH and maximum shear stress. (The shear stress data are shown in Figures 3c and 4c). The pH of each flake was measured using a needle point pH probe on the raw fillet and matched with the textural data for that flake after the fillet was cooked. The pH values varied considerably within a region and only within the lower, central region was there any correlation with texture measurements. The pH values changed very little between regions (the tough dorsal region had approximately the same pH as the more tender central region) and thus when all the pH-texture data for this fillet is considered, the correlation is very poor. The average pH of a fish tells a great deal about the overall textural quality and variability, but more precise pH measurements in terms of location and numbers adds no new information.

## CONCLUSIONS

1. The large variations that occur within a fish or fillet make sample randomization a poor choice for studies of storage conditions, process variables, or sensory-instrumental correlations. Careful selection of uniform regions provides a better alternative.

2. The variations within fillets appear predictable.
   a. Low pH fish are tougher and more variable than high pH fish.
   b. Dorsal section of the fillet will be tougher than other sections.
   c. The mid-length of the fillet tends to be slightly more tender than the ends, particularly the head end of the fillet.

3. Large regions of uniform textural properties can be identified in most fillets. In most cases only 30-40% of the fillet will be wasted and much of this wasted material would not be used in studies where samples are randomized. In general, the central section from the 10th to the 25th flake is the most uniform section.

4. The average pH of the fillet correlates well with the overall textural quality of the fillet and with the overall textural variation. However, the pH variations within the fillets do not correlate with the textural variations within the fillets.

## REFERENCES


1. Kapsalis, J.G. Consumer and Instrumental Edibility Measures for Grouping of Fish Species, NTIS, 1980.

2. Love, R.M. and I. Robertson. The connective tissues of fish. I. the influence of biological condition in cod on gaping in frozen-thawed muscle. Journal of Food Technology 3215-221. 1968

3. Love, R.M. Variability in Atlantic Cod (Gadus morhua)

from the Northest Atlantic: a Review of Seasonal and Environmental Influences on Various Attributes of the Flesh. Journal Fisheries Research Board of Canada, 32 (12): 2333-2342. 1975.

4. Segars, R.A., R. Hamel, J. Kapsalis. A Punch and Die Test Cell for Determining the Textural Quality of Meat. Journal of Texture Studies 6: 211-225. 1975.

# PRELIMINARY COMPARISON OF PASTEURIZATION METHODS FOR FLAKED FISH

Melvin E. Waters
National Marine Fisheries Service, NOAA
Southeast Fisheries Center, Charleston Laboratory
Charleston, South Carolina 29412-0607

## Introduction

One primary objective of technologists when developing methods to preserve or extend the shelflife of perishable foods is to maintain, as nearly as possible, the original "fresh-like" characteristics of the product. Most seasonally harvested foods require some method of preservation to maintain a degree of off-season availability. For the most part, consumers prefer to purchase their food in the fresh state, a fact evidenced by elevated sales during the season of harvest. The next choice is pasteurized foods, followed by frozen, canned, smoked, dried and salted foods. Frozen foods are expensive to prepare and maintain during storage and marketing and, seafoods in particular, are highly susceptible to undesirable quality changes even for short periods of storage. Conversely, canned foods remain acceptable after long periods of storage and can be stored at room temperature. However, thermal processing at 115 and 120°C causes large texture changes and imparts special canned odors and flavors to the product. In addition, nutrient properties of proteins are severely damaged. Amino acids such as tryptophane, methionine and others are partially destroyed and the net protein value of heat processed products is about 75 to 80 percent (Wierbicki, 1982) to that of the unheated product. Smoked, dried and salted products require special care during storage and many of the product attributes are destroyed by the process.

Pasteurized foods are highly desirable because minimal changes take place in the product during processing and the resulting product more nearly resembles that of the fresh state. Pasteurized products, however, must be stored at refrigerated temperatures (2-3°C) and have a shelflife ranging from a few days to about 6 months. Foods pasteurized in hermetically sealed containers offer several of the preferable product characteristics of other preservation methods and provides a product highly acceptable to the consumer.

Thermal pasteurization of foods is a long standing process. The pasteurization of milk, wine, cheese, etc., has been practiced for years and these products have been fully accepted by consumers. Pasteurization has a broad meaning in modern technology of food processing. In this sense it means a heat treatment of foods that is intended to destroy vegetative forms of pathogens or that which will destroy all or most vegetative forms of organisms that cause spoilage or that interfere with desirable fermentations.

The heat pasteurization of fishery products is limited almost entirely to that of crab meat. Byrd (1951) patented a process that extends the shelflife of crab meat to about 6 months, retaining many of the properties of the cooked-steamed product. The process involves the heating of hermetically sealed cans of fresh crab meat to an internal temperature of 88°C for about 1 minute, followed with rapid cooling and subsequent storage at 1 to 4°C. The process is being used commercially and the product appears to be well accepted by the consumer. Commercial pasteurization of crab meat is a time-consuming and, for the most part, a batch process that must be carefully controlled to ensure proper heating of all containers.

Contribution No. 82-15C, Southeast Fisheries Center, Charleston Laboratory

Littleford (1957) found that 76 minutes were required to heat 211 X 400 cans of crab meat to an internal temperature of 76°C in a 79°C water bath starting with an initial temperature of about 1°C.; 47 minutes were required to reach an internal temperature of 76°C in a 90°C water bath. He further showed that nothing will be gained with respect to shelflife and bacteriological acceptability by extending the holding period at 76°C (internal temperature) beyond 1 minute. Shorter exposure time at pasteurization temperatures may further preserve the "fresh-like" characteristics of seafoods but, more importantly, would reduce processing cost (labor, utilities, etc.). Microwave (MW) processing may be an economical and effective alternate method to pasteurize fishery products.

The use of MW energy in the preparation of foods is becoming more prevalent in homes, institutions and businesses, principally for its rapid heating properties. This cooking process generally retains good product texture and flavor and consumes less energy than conventional methods. Because MW cooking apparently has advantages over conventional cooking, its effectiveness in reducing food-borne organisms needs to be considered.

Research demonstrating the application and effectiveness of MW energy in the preparation and preservation of foods has been reported in the literature (Cunningham and Francis, 1982; Wu and Salunkhe, 1977; Ayoub et al., 1974; Craven and Lillard, 1974; Kenyon et al., 1971; Mendelsohn et al., 1969). The research shows that the process can be used successfully in a variety of applications, running the gamut from shucking oysters to the sterilization of meat and poultry. Microwave heating is moving rapidly from the laboratory and pilot-plant into industrial processing with several systems now in operation.

Microwave pasteurization of fishery products has not been reported in the literature. However, some preliminary studies using crab meat were conducted in 1970 by Dr. Mahlon C. Tatro, University of Maryland. Dr. Tatro experienced several problems and abandoned the work without publishing his results. The problems included uneven temperature distribution within containers, rupturing of the containers due to a build-up of pressure and arcing of thermocouples placed in containers to monitor the temperature during the heating process.

The MW pasteurization of fish flakes appears to be a feasible process and would be beneficial to the seafood industry at a time when they are seeking new opportunities to market underutilized species and looking for ways to reduce production costs. The objectives of this study were to: (1) determine the effectiveness of MW pasteurization in extending the shelflife of crab meat and fish flakes, (2) determine the affect of MW pasteurization on sensory properties of fish flakes, (3) approximate the time required to heat containerized fish flakes to pasteurization temperatures using MW energy and (4) to determine temperature variability within individual containers of MW pasteurized fish flakes.

## Materials and Methods

Pasteurization Equipment:

A Sharp[2/] household-model MW oven (2450 MHz, 1.5 kilowatts power output), equipped with a carousel, was used as the source of MW energy. A retort filled with water and maintained at 90°C with an inflow of steam was used to duplicate commercial pasteurization methods.

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2/ The use of trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

Product Preparation:

Fresh, iced mullet (Mugil cephalus) were obtained from a local seafood dealer approximately 36 hours after harvest. Iced red porgy (Pagrus pagrus) were obtained from the South Carolina R/V Atlantic Sun and were harvested approximately 48 hours before preparation. Mullet and red porgy were scaled, headed, gutted and thermocouples placed in the geometric center of six fish. The fish were steam-cooked 15-20 minutes under 5 pounds of pressure and held overnight in a cooler. The flesh was removed from the skin and bones and separated into flakes. Freshly picked, flaked crab meat (Callinectes sapidus) was obtained from a processor in Beaufort, S.C., packed in ice and transported to the laboratory for processing.

The three flaked products were each divided approximately in half; one half of each product was packed in high-density polyethylene containers and the other in 307 X 113 C-enamel cans. One hundred and twenty grams of flesh were packed in each of the polyethylene containers and the lids attached loosely; 200 grams were placed in each of the cans and the cans sealed at room temperature. Thermocouples were placed in six cans of flaked products to monitor the temperature during pasteurization. No attempt was made to monitor the temperature of polyethylene containers of flakes during MW heating.

Pasteurization Process:

A preliminary experiment was conducted to establish MW pasteurization parameters. Three polyethylene containers of each product were heated 5, 7, 9 and 12 minutes in the microwave oven and the temperature monitored with thermocouples at several locations in each container for 5 minutes after removal from the oven. The experiment was repeated confirming a process temperature/time relationship of 80°C when heated 9 minutes .

Products packed in polyethylene containers were pasteurized by MW energy and cans were pasteurized in a water bath (WB). Water-bath pasteurized flakes served as the control. Twenty polyethylene containers of each product were placed in the MW oven, lids loosely attached, and heated 9 minutes. The flaked products were removed from the oven, allowed to equilibrate 5 minutes and the lids snapped into place providing air-tight containers. Cans of flaked products were placed in a 90°C WB and heated to an internal temperature of 85°C. All products were then stored in a cooler at 2.5°C.

Product Evaluation:

Before pasteurization, three containers (two polyethylene containers and one can) of each product were analyzed organoleptically, microbiologically and chemically to establish a base for determining the effectiveness of each process. After processing, three containers representing each product and each process were again evaluated microbiologically, organoleptically and chemically after 0, 1, 3 and 6 months of storage.

Microbial analyses -- Microbial samples were obtained aseptically from containers before conducting sensory and chemical analyses. Total aerobic plate counts (TAPC) were determined according to the procedure outlined in FDA's Bacteriological Analytical Manual for Foods (Association of Official Analytical Chemists, 1976). Standard plate count (SPC) agar was used as the

plating medium and plates were incubated 72 hours at 35°C. Proteolytic microorganisms were determined using SPC agar fortified with skim milk (2%) and plates incubated 72 hours at 35°C. Colonies surrounded by a clear zone were counted as proteolytic organisms.

Sensory evaluations -- The color, odor and general appearance were assessed informally by three panelists experienced in judging the quality of processed fishery products.

Chemical analyses -- Total volatile nitrogen (TVN), trimethyl-amine-nitrogen (TMA-N), moisture and pH were measured on samples passed through a meat grinder three times to obtain a homogeneous mixture. TVN and TMA-N content were determined as described by Cobb et al. (1973); moisture and pH analyses were conducted using AOAC methods (1975).

Microbial and chemical values are reported as an average of three analyses.

## Results and Discussion

The weights of whole mullet used in this study were 600 to 700 grams each. When cooked in the retort 15 minutes under 5 pounds pressure, the fish reached an internal temperature of 88°C. The yield of flaked meat (based on whole weight) was 32 percent and the moisture content 66 percent. The weights of whole red porgies were 1,000 to 1,200 grams each and required 20 minutes steaming under 5 pounds pressure to reach an internal temperature of 88°C. The yield of flaked meat was 36 percent and the moisture content was 68 percent. Commercially picked crab meat contained 76 percent moisture.

In the preliminary experiment with flaked fish to establish processing parameters for MW heating, the temperature varied considerably throughout the container. The temperature was lowest at center-top and highest at center-bottom with temperatures in between at mid-center and for various locations along the sides. The temperature varied as much as 15°C between top and bottom when measured immediately after removal from the oven. However, within 5 minutes after removal from the oven, the temperature at the cooler points increased and began to equilibrate with the higher temperatures. At 5 minutes post pasteurization, the temperature varied less than 5°C throughout the container and the temperature at all points began to decrease. Variability in heat distribution within MW ovens has been reported by other investigators (Baldwin et al., 1971; Lacey and Winner, 1965).

Cans of flaked products with an initial temperature of 15°C and heated in a WB at 90°C, required 45 minutes processing time to reach an internal temperature of 85°C.

Results of the chemical analyses of pre-pasteurized, pasteurized and stored flakes are shown in Figure 1. Analyses of the variance (ANOV), at the 95% confidence level, were applied to the chemical data to determine if significant differences exist between pre-pasteurized, MW- and WB-pasteurized flakes at 0-months of storage and between MW- and WB-pasteurized flakes at 1, 3 and 6 months of storage. Analyses of the data on mullet and red porgy revealed that there were significant differences in pH and TVN values for all treatments at all levels of storage. TMA-N values for mullet were significantly different except at 3 months of storage. TMA-N values for red porgy were significantly different between treatments at 0 and 1 month of storage but not significantly different at 3 and 6 months. TMA-N content is indicative of activity of some common spoilage bacteria in seafood products.

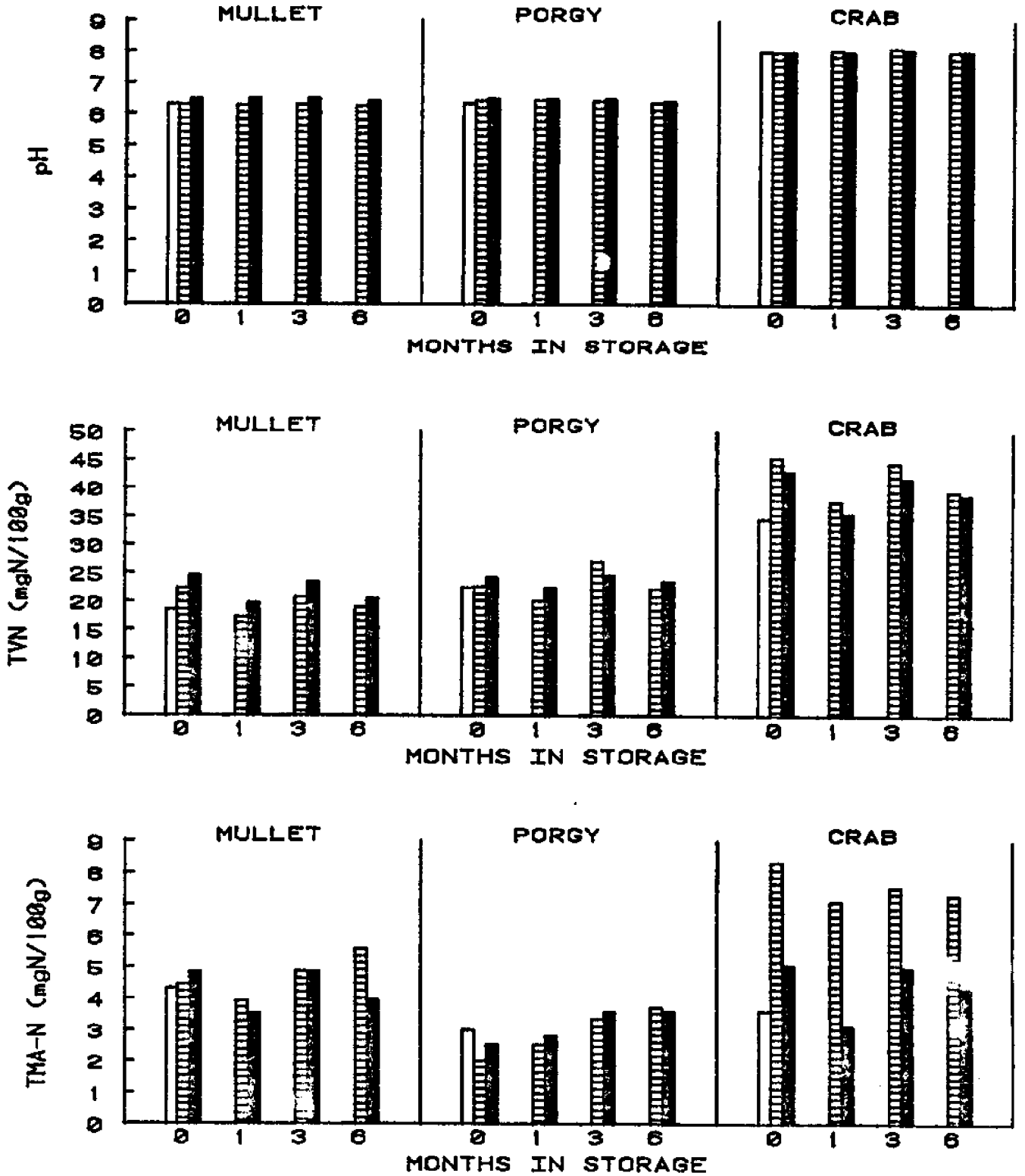


Figure 1. Graphic representation of chemical values for pre-pasteurized (□), microwave - pasteurized (▤), and water bath - pasteurized (■) fish flakes and crab meat stored at 2.5°C.

Analysis of the data on crab meat showed a significant difference between pH values at 0 months of storage, TVN values at 0, 1 and 3 months and TMA-N values at all levels of storage. Values for all quality attributes were greater for crab meat than for mullet or red porgy.

Observation of Figure 1 reveals that only small differences exist between heat treatments for most quality factors and for all products. Although some statistically significant differences exist, these differences may not have practical significance. Generally speaking, and as expected, chemical indices did not increase during storage due to the inactivation of bacterial and autolytic enzymes during the pre-cooking process and to low bacterial counts, as will be discussed later in this paper.

The microbial data, summarized in Table 1, demonstrates that pasteurization effectively reduced the bacterial counts for all products evaluated. This data was not analyzed statistically because of the large variability in counts among treatments and storage periods. Both pasteurization methods substantially reduced the TAPC and totally eliminated proteolytic organisms. Although the TAPC of products pasteurized by both methods varied during storage, the counts did not increase substantially. Low initial microbial counts for mullet and red porgy flakes are attributed to preparation using good sanitary practices. The high initial count of crab meat was probably due to hand processing and/or improper storage of the crab meat before and after handpicking. The high initial count is likely responsible for the high pH, TVN and TMA-N content as illustrated in Figure 1. The elimination of proteolytic microorganisms by pasteurization correlated well with the stability of chemical values (quality indices) during storage. The MW-pasteurization method was as effective as the control method in reducing the microbial count.

Sensory panelists rated the WB-pasteurized (control) mullet and red porgy flakes slightly higher than the MW-pasteurized flakes. A slight orange-brown discoloration formed on the surface of the MW-pasteurized flakes and some surface dehydration was visible, accounting for the lower scores. The discoloration and dehydration was probably due to containers not being completely sealed allowing moisture loss.

The panelists rated the control and the MW-pasteurized crab meat borderline for general appearance. A blue-grey discoloration formed in both pasteurized products, similiar to that occasionally found in canned crab meat, and has been related to the product pH. Waters (1971) showed that treating crab meat with a phosphate/citric acid buffer solution reduced the pH and prevented blue discoloration in the canned product. The control appeared very moist while the MW-processed crab meat was less moist. All other products were extremely dry, suggesting excessive precooking. Post-processed products were not analyzed for moisture content but this may be an important factor in establishing microwave processing parameters. Mr. R.E. Mudgett, University of Massachusetts, stated at the recent Institute of Food Technologist Meeting in New Orleans, Louisiana, "depth of penetration of microwaves is much greater for materials with lower moisture content." The relationship between the moisture content and MW penetration of foods should be investigated. No spoilage odors were reported by the sensory panel from any of the products during storage.

Table 1. Results of the microbial analysis of pre-pasteurized, microwave - and water bath - pasteurized fish flakes and crab meat.

| PASTEURIZATION METHOD | MONTHS IN STORAGE | MICROBIAL COUNTS (Count/g) | | | | | |
|---|---|---|---|---|---|---|---|
| | | MULLET | | RED PORGIE | | CRAB MEAT | |
| | | TAPC[1/] | PROTEO.[1/] | TAPC[1/] | PROTEO.[1/] | TAPC[1/] | PROTEO.[1/] |
| Pre-Pasteurization | | 4,000 | 100 | 5,700 | 200 | 1,100,000 | 3,200 |
| Microwave | 0 | 100 | 0 | <100 | 0 | 200 | 0 |
| | 1 | 700 | 0 | 1,000 | 0 | 200 | 0 |
| | 3 | 100 | 0 | <100 | 0 | 100 | 0 |
| | 6 | 1,800 | 0 | 3,300 | 0 | 200 | 0 |
| Water Bath (Controls) | 0 | <100 | 0 | 700 | 0 | 100 | 0 |
| | 1 | 1,000 | 0 | 1,000 | 0 | 300 | 0 |
| | 3 | <100 | 0 | <100 | 0 | <100 | 0 |
| | 6 | 200 | 0 | <100 | 0 | 500 | 0 |

1/ TAPC = Total Aerobic Plate Count

Proteo. = Proteolytic

Results obtained from this study showed that MW-pasteurization of containerized fish flakes and freshly picked crab meat to an internal temperature of 80°C is an effective and feasible process. Craven and Lillard (1974) reported that MW heating completely destroyed Clostridium perfringens 7947 cells when internal temperatures exceeded 70°C in chicken innoculated with $10^5$ cells/gram or when internal temperatures exceeded 64°C in chicken innoculated with $10^3$ cells/gram. Baldwin et al. (1971) showed that MW heating of innoculated tuna pies and casseroles, fish fillets and fish sticks completely destroyed Salmonella typhimurium when heated approximately 7 minutes to an internal temperature of 55°C. Wu and Salunkhe (1977) showed that MW heating of protein curds to 80°C, and subsequent storage at 4.5°C, extended the shelflife from 7 days for the control to 21 days for the MW treatment. Cunningham and Francis (1982) demonstrated that 20 to 30 seconds exposure of fresh poultry pieces to MW heating significantly reduced the bacterial count after 7 days of storage at 4°C.

Our results indicate that the refrigerated shelflife of fish flakes and crab meat can be extended to 6 months with MW-pasteurization and that the process is as effective as the conventional WB method. The problems encountered in our work regarding discoloration and dehydration are minor and can be easily solved with additional effort. The MW heating period of 9 minutes appears excessive and may have contributed to the discoloration problem; the exposure time should be reevaluated.

It was not the intent of this work to investigate mechanical problems involved in MW processing. The problem of ruptured containers during MW heating has already been resolved and demonstrated by the development of a continuous sterilization system utilizing external air pressure in the range of 30 to 45 psig (Ayoub et al., 1974; Kenyon et al., 1971). Reinforced flexible pouches and plastic containers are available to minimize the problem. The lack of uniform heat distribution in MW heated products has been minimized through the use of mechanisms such as turntables or belts to move containers of the product during processing.

The results of this preliminary study were intended to provide basic information in the development of alternate methods for the preservation of fish flakes. Economics of MW-pasteurization were not determined but savings in energy and labor costs are obvious. Additional work is required before the process can be recommended as a safe commercial process because of the possible survival of Clostridium botulinum spores. The results do indicate that the development of pasteurized fish flakes as a product parallel to commercially pasteurized crab meat is a promising approach in utilization of southeastern fishery resources.

## Conclusion

Microwave pasteurization effectively extends the shelflife of laboratory prepared fish flakes and fresh-picked crab meat up to 6 months when stored at 2.5°C. Microwave pasteurization is as effective as the conventional, water bath, pasteurization method and appears to be more economical. Extended exposure of fish flakes to MW for 9 minutes may have caused some discoloration of products and exposure time should be reevaluated. Temperature variations within individual containers of MW-pasteurized flakes tended to equilibrate within 5 minutes after processing.

## REFERENCES


Association of Official Analytical Chemists. 1975. Official Methods of Analysis, 12th ed. Association of Official Analytical Chemists, Washington, D.C.

Association of Official Analytical Chemists. 1976. Bacteriological Analytical Manual for Foods. Association of Official Analytical Chemists, Washington, D.C.

Ayoub, J.A., D. Berkowitz, E.M. Kenyon and C.K. Wadsworth. 1974. Continuous microwave sterilization of meat in flexible pouches. J. Food Sci. 39:309-313.

Baldwin, R.E., M.L. Fields, W.C. Poon and B. Korschgen. 1971. Destruction of Salmonella by microwave heating of fish with implications for fish products. J. Milk Food Technol. 34:467-470.

Byrd, G.C. 1951. Method of keeping the meat of shellfish in a fresh condition. United States Patent Office, Pat. No. 2,546,428.

Cobb, B.F. III, I. Alaniz and C.A. Thompson, Jr. 1973. Biochemical and microbial studies on shrimp: volatile nitrogen and amino nitrogen analysis. J. Food Sci. 38:431-436.

Craven, S.E. and H.S. Lillard. 1974. Effect of microwave heating of precooked chicken on Clostridium perfringens. J. Food Sci. 39:211-212.

Cunningham, F.E. and Carol Francis. 1982. Use of microwaves to extend shelflife of refrigerated poultry. Feedstuffs 54(2):23-24.

Kenyon, E.M., D.E. Westcott, P. LaCasse and J.W. Gould. 1971. A system for continuous thermal processing food pouches using microwave energy. J. Food Sci. 36:289-293.

Lacey, B.A. and H.I. Winner. 1965. Effects of microwave cookery on the bacterial counts of foods. J. Appl. Bacteriol. 38:331-335.

Littleford, R.A., 1957. Studies on pasteurization of crabmeat. Bulletin No. 2, July 1957, University of Maryland Seafood Processing Laboratory, Crisfield, Maryland, p. 14.

Mendelsohn, J.M., L.J. Ronsivalli, F.J. King, J.H. Carver, R.J. Learson, B.W. Spracklin and E.M. Kenyon. 1969. Opening oysters and other bivalves using microwave energy. Fish. Ind. Res. 4(7):241-248.

Waters, M.E. 1971. Blueing of processed crab meat. II. Identification of some factors involved in the blue discoloration of canned crab meat (Callinectes sapidus). Special Scientific Report - Fisheries No. 633.

Wierbicki, E. 1982. Shelf stable irradiated meat. Research on wholesomeness lays groundwork for future development. Food Development 16(1):24-28.

Wu, M.T. and D.K. Salunkhe. 1977. Extending shelf-life of fresh soybean curds by in-package microwave treatments. J. Food Sci. 42:1448-1450.



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# ENZYMATIC AMMONIA AND UREA DETERMINATIONS IN FRESH SEAFOODS HELD REFRIGERATED AND ON ICE

W.L. Cheuk, G. Finne, and R. Nickelson
Seafood Technology
Texas A&M University
College Station, Texas 77843

## INTRODUCTION

Quality changes which occur in fresh seafood products during refrigerated or iced storage are the combined results of both microbiological and tissue enzyme activities. Both give rise to a number of different volatile compounds of which ammonia is the most important. Due to the build-up of ammonia during storage of fresh seafoods, its concentration has often been suggested as an objective index of fresh quality. Ward et al., (18) using an ammonia specific electrode, demonstrated the relationship between storage time, total microbial numbers and ammonia concentration during refrigerated storage of fresh shrimp. Ammonia has also been suggested as an index of quality for crabmeat (3, 15) dogfish and thornback ray (17). For elasmobranch species (sharks, dogfish, rays and skates), post-mortem ammonia production is especially a problem. These fishes contain high levels of urea both in blood and tissue, and during iced or refrigerated storage urease positive microorganisms will rapidly convert urea to ammonia. Both France and Belgium, where dogfish is used as a foodfish, have stringent quality standards based on the ammonia content of this species.

A number of quantitative methods for the determination of ammonia in seafood products have been developed. Most of these methods rely on microdiffusion, distillation and/or ion exchange. After being liberated from the tissue by strong alkali, ammonia is trapped in weak acids and measured titrimetrically (6) or photometrically by either the Nessler's reagent (13) or the Berthelot reaction (4).

When researching the kinetics of conversion of urea to ammonia in sharks held on ice, we adapted an enzymatic method for the simultaneous determination of urea and ammonia. The method is based on the determination of ammonia in body fluids and tissue as reported by Faqaz and Dahl ( 7) and later by Mondzac et al., (12). Knight and Toom (10) showed that the method was well suited for the determination of ammonia in seafoods. They also demonstrated that, unlike other methods, the enzymatic technique was specific for ammonia and other decomposition products and food additives did not interfere with the assay. Talke and Schubert (16) coupled the enzymatic ammonia determination in blood with urease thus providing a convenient way to measure both ammonia and urea in the same aliquot in a single step. We have found that when adapting this

method to seafood products both ammonia and urea can be determined accurately with good reproducibility.

## METHODS

### Appratus

(a) Spectrophotometer. - Perkin-Elmer, Model 124, double beam grating spectrophotometer with automatic recorder. Place slit selector at 0.5 nm, which, according to operation manual, provides a bandpass of 0.5 nm.

(b) Cuvettes - 1 - cm lightpaths, square quartz curvettes capable of holding 3.5 ml volume.

(c) Centrifuge - Damon/IEC refrigerated centrifuge Model B-20A.

### Reagents

(a) Buffer - (Triethanolamine, 0.5 mol/l; α-ketoglutarate, 35 mmol/l, pH-8.6). Dissolve 9.3 g triethanolamine hydrochloride (Boehringer Mannheim Biochemicals, Indianapolis, IN) and 670 mg α-ketoglutarate disodiumsalt (BMB) in 70 ml distilled water, adjusted to pH 8.6 with sodium hydroxide (Fisher Scientific, Fair Lawn, NJ), 5 mol/l, and fill up to 100 ml with distilled water. The buffer is stable for at least 4 weeks at 4°C.

(b) Reduced nicotinamide adenine dinucleotide solution (NADH) 6 mmol/l. Dissolve 30 mg NADH-$Na_2$ (BMB) and 60 mg sodium bicarbonate in 6 ml distilled water. Stable for 4 weeks at 4°C.

(c) Glutamate dehydrogenase (GLDH), 10 g/l. - Commercial GLDH in glycerol solutions (BMB) can be used undiluted. Stable for at least 1 year at 4°C.

(d) Urease, 2.5 g/l. - Dissolve 10 mg lyophilisate urease (available as 2.5 mg enzyme protein (BMB) in 1 ml glycerol (50%, v/v). Stable for 4 weeks at 4°C.

(e) Perchloric acid, 1 mol/l. - Pipette 15 ml 70% perchloic acid (Fisher Scientific, Fair Lawn, NJ) into a 100 ml flask with 60 ml of distilled water, bring to volume.

(f) Potassium hydroxide, 2 mol/l. - Dissolve 11.22 g potassium hydroxide (Fisher Scientific, Fair Lawn, NJ) in 100 ml distilled water.

### Seafood Samples Tested

Tails of brown shrimp (Penaeus aztecus), fillets of flounder (Paralichthys lethostigma) and freshly picked crabmeat (Callinectes sapidus) obtained from a seafood retail store in Aransas Pass, Texas, were packed on ice and shipped to the laboratory in College Station, Texas. The shrimp tails were stored refrigerated at 3.5°C, the crabmeat in plastic bags on ice and the flounder fillets also on ice. At regular intervals during the storage period samples were analyzed for

ammonia and urea by the enzymatic assay, total volatile nitrogen (TVN), trimethylamine (TMA) (5) and aerobic plate count (APC) (1).

Preparation of Samples

The extraction procedure and assay conditions used during this study were similar to those described in Methods of Enzymatic Food Analysis (2).

Extract 10 g sample with 20 ml perchloric acid for 5 min in a blender or homogenizer. Centrifuge at 5,000 rpm for 15 min. Pipette 10.0 ml of the supernatant into a beaker and neutralize with the potassium hydroxide solution. Record volume of KOH added. Store the mixture in a refrigerator for at least 10 min, filter and dilute (if necessary) the clear filtrate and take 0.1 ml of the enzymatic assay.

Determination

The reaction scheme for the enzymatic determination of ammonia and urea is given in equation 1 and 2.

Equation 1:

$$\text{urea} + H_2O \xrightarrow{\text{urease}} 2\ NH_3 + CO_2$$

Equation 2:

$$NH_4^+ + \alpha\text{-ketoglutarate} + NADH \xrightarrow{GLDH} L\text{-glutamate} + NAD^+ + H_2O$$

Abbreviations:

GLDH - L-glutamate dehydrogenase

$NAD^+$ - Nicotinamide Adenine Dinucleotide

NADH - Nicotinamide Adenine Dinucleotide, Reduced

(a) Enzymatic assay

Ammonia. Pipette 1.00 ml buffer, 0.10 ml NADH solution, 0.10 ml sample and 1.90 ml distilled water into the quartz cuvette. Mix thoroughly. After approximately 3 min, read absorbance of the mixture against water at 340 nm ($A_1$ ammonia). Start the reaction by the addition of 0.02 ml GLDH solution (Equation 2), mix well and allow reaction to be completed (15 to 25 min). Read absorbance ($A_2$ ammonia). If the reaction is still running after 25 min, continue to read absorbance at 2 min intervals until constant rate. The ammonia concentration is calculated from absorbance difference between $A_1$ and $A_2$ or from constant rate graph shown in Figure 1.

For the determination of urea, continue the reaction by the addition of 0.02 ml urease solution, mix well and proceed as for ammonia. Note final absorbance ($A_3$) and calculate urea from difference between $A_2$ and $A_3$ ($\Delta$ A urea) or as shown in Figure 1.

(b) Calculations

The concentration of ammonia or urea in the assay solution can be calculated according to equation:

$$c = \frac{V \times EW \times \Delta A}{E \times d \times v \times 1000} \quad (g/l)$$

where V = final cuvette volume (ml)

v = sample cuvette volume (ml)

EW = equivalent weight of substance to be assayed

d = lightpath (cm)

E = extinction coefficient of NADH at 340 nm. (6.3 liter $mmol^{-1}cm^{-1}$)

c = concentration

When calculating the concentration of ammonia and urea in the samples, the quantity of moisture in the sample must be taken into consideration. If the sample weight is 10 g with a moisture content of 78%, 7.8 ml moisture is added to sample liquid volume after protein denaturation. A moisture content of 78% is a good average for fresh seafoods.

Calculation example:

Ammonia in flounder at 0 day storage.

| | | |
|---|---|---|
| Weight of sample $W_s$ | = | 10.0 g |
| KOH used to neutralize | = | 6.9 ml |
| Total volume (20 ml $HClO_4$ + 7.8 ml $H_2O$) | $V_1$ = | 27.8 ml |
| Volume of supernatent | $V_2$ = | 10.0 ml |
| Volume of sample mixture ($V_2$ + KOH used) | $V_3$ = | 16.9 ml |
| Volume taken from sample | = | 0.2 ml |
| Absorbance difference | = | 0.81 OD |

$$\% \text{ Ammonia} = (\text{g } NH_3/1) \times \frac{V_1 \times V_3 \times 100}{V_2 \times W_x \times 1000}$$

$$= \frac{0.81 \times 3.22 \times 17.03}{6.3 \times 1 \times 0.20 \times 1000} \times \frac{27.8 \times 16.9 \times 100}{10 \times 10 \times 1000}$$

$$= \underline{0.0\ 1653\%} \quad \text{or} \quad \underline{16.53 \text{ mg}/100\text{g}}$$

Instead of using the extinction coefficient of NADH to calculate ammonia and auea, a standard curve showing the absorbance difference at different concentrations can also be used. A standard curve for urea is prepared as follows: Accurately weigh 30 mg analytical grade urea (Fisher Scientific, Fair Lawn, NJ) into a 100 ml volumetric flask and bring to volume with distilled water. Dilute this stock solution into a series of appropriate concentrations. Perform the enzymatic assay for each dilution and construct a standard curve showing change in absorbance at 340 nm for each concentration used. A standard curve for ammonia can also be constructed from alnalytical grade ammonium salts.

## RESULTS AND DISCUSSION

The direct enzymatic determination of ammonia and urea have a number of advantages over other methods commonly used. The primary advantage is that both compounds can be determined in the same sample using the same technique. A secondary advantage lies in the specificity of the enzymes (12, 10). While total volatile nitrogen analysis will include a number of amines in addition to ammonia, this enzymatic assay is specific for either ammonia or urea. Except for a narrow band width UV spectrophotometer, no special appratus are needed.

After the sample has been deprotenized by perchloric acid all production of ammonia and urea is arrested. There is however, a potential source of interference of a low rate liberation of ammonia during the assay. The neutralized supernatant after protein precipitation may contain short peptides or free amino acids from which ammonia may be slowly released in the alkaline buffer (pH 8.6). During this study, this side reaction was seldom encountered. By using the extrapolation technique as described in Figure 1, this problem when occurring can be easily eliminated.

In order to test the applicability of the enzymatic determination for ammonia and urea in seafoods, fresh shrimp tails, crabmeat and flounder fillets of various degrees of decomposition were analyzed for urea and ammonia together with traditional spoilage tests. Table 1 shows the concentrations of ammonia, urea, total volatile nitrogen, trimethylamine and total aerobic plate count of shrimp tails stored refrigerated at 3.5°C. There was a close relationship between the different spoilage parameters. Using TVN as an index, Stansby (14) listed

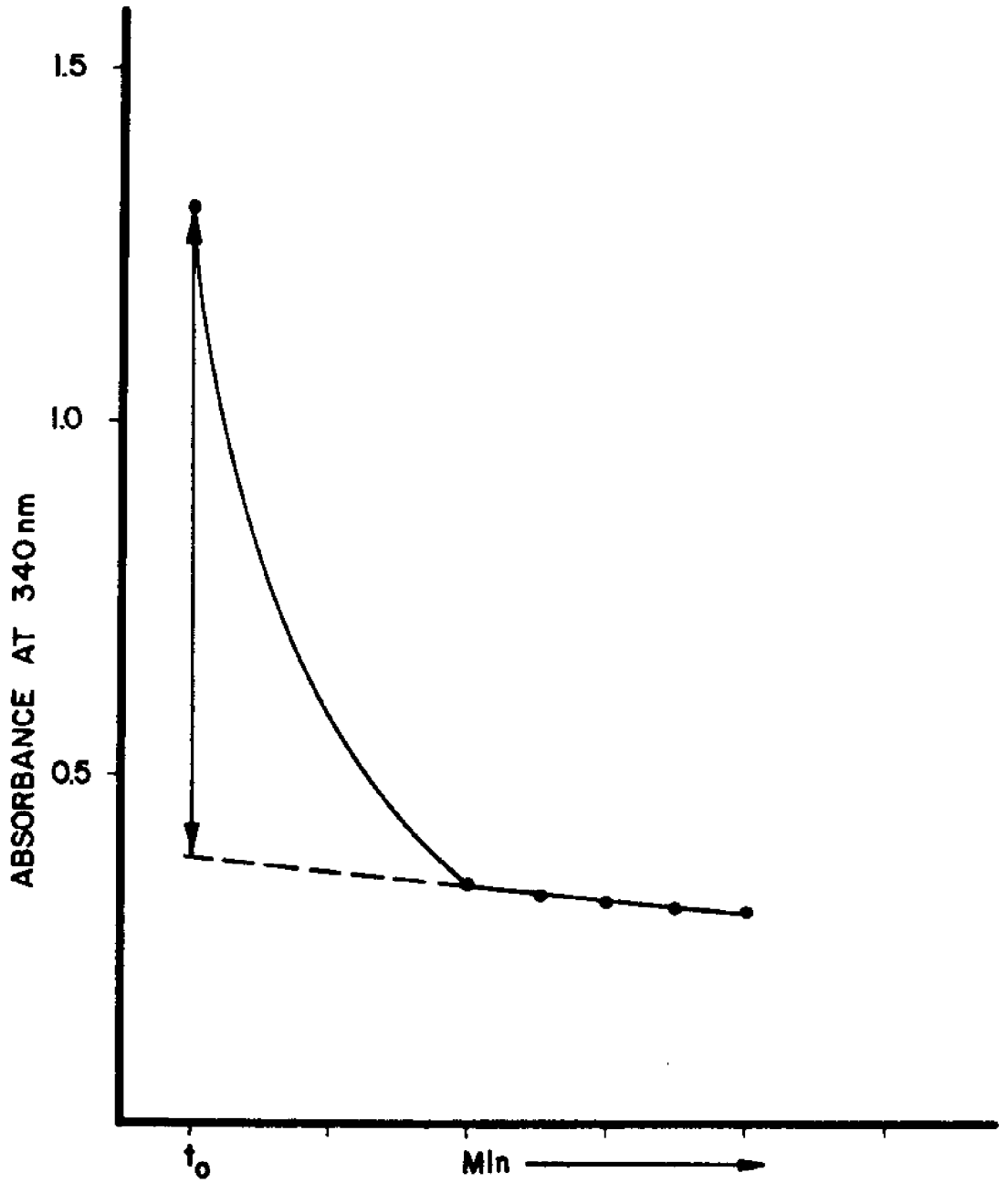


Figure 1. Graphical extrapolation of absorbance during enzymatic assay.

| Day | Urea (mg/100g) | Ammonia (mg/100g) | mg TVN-N/100g | mg TMA-N/100g | Aerobic Plate Count |
|---|---|---|---|---|---|
| 1 | 0.74 | 4.92 | 7.51 | traces | $1.49 \times 10^5$ |
| 3 | 1.05 | 10.35 | 8.34 | traces | $1.16 \times 10^6$ |
| 5 | 1.38 | 18.09 | 15.42 | 2.45 | $3.4 \times 10^6$ |
| 7 | 2.43 | 23.98 | 28.79 | 4.52 | $1.69 \times 10^7$ |
| 9 | 2.70 | 40.05 | 43.40 | 7.82 | $2.29 \times 10^7$ |

Table 1. Ammonia, urea, total volatile nitrogen (TVN), trimethylamine (TMA) and aerobic plate count (APC) of shrimp tails stored at 3.5°C.*

* All data were the average of two samples.

the following values (mg TVN-N/100 g) for different degrees of freshness of fish; 12 or less, fresh fish; 12-20, edible with only slight decomposition; 20-25, borderline; and above 25, inedible and decomposed. Even though such a classification may not be directly applicable to shrimp, a value of 7.51 TVN-N/100g indicated good initial quality. During storage at 3.5°C, ammonia and urea increased as the total plate count and the traditional spoilage indicators increased. A close relationship between ammonia and TVN was expected since Cobb et al. (5) showed ammonia to be the principal component of the total volatile nitrogen fraction in shrimp held on ice. When using a TVN value of 25 mg TNV-N/100g as an indicator of inedibility (14), the shrimp used for this study were spoiled after 7 days at 3.5°C. At this point the aerobic plate count had reached $1.69 \times 10^7$/g, the TVN value was 28.79 mg TVN-N/100 g and the ammonia concentration was 23.98 mg $NH_3$/100 g. The urea content in shrimp tails also increased during fresh storage which is in agreement with Cobb et al. (5).

The ammonia, urea and TVN content of flounder fillets held on ice showed an initial decrease during ice storage (Table 2). Such decreases in water soluble compounds used as quality indicators when holding fresh seafoods on ice have been reported in earlier studies (9) and is believed to be due to the leaching action of the melting ice. After seven days unprotected on ice, the fillets were put in polyethylene plastic bags and placed back on the ice. When stored in a protective barrier, the trend was immediately reversed and ammonia, urea and total volatile nitrogen compounds increased sharply.

On the basis of organoleptic evaluations, Hillig et al., (8) defined three stages of decomposition for fresh crabmeat. In a later collaboratory study, Steinbrecher (15) showed statistically significant differences in ammonia between the three stages and recommended that ammonia concentration be adopted as an index of quality. Table 3 shows the development of ammonia and other quality parameters during storage of fresh crabmeat. The most noticeable difference between the crabmeat and the other fresh seafoods tested was the dramatic increase in urea during storage. Since fresh crabmeat is a cooked product, endogenous enzymes associated with the urea cycle cannot explain the build-up of urea during storage. The high ammonia content and rapid spoilage of fresh crabmeat is most likely due to the invasion of urea positive psychrotrophic microorganisms which will convert urea to ammonia. The ammonia content of the crabmeat used in this study was initially 41.73 mg $NH_3$/100 g. Using a value of 45 mg $NH_3$/100 g as a cut-off point as suggusted by Steinbrecher (15), the initial quality fell within Class 1. However, spoilage proceeded rapidly and after only six days on ice the crabmeat has reached Class 3 quality (94.3 mg $NH_3$/100 g) at this point the microbial population was more than $10^8$ organisms/g.

This study has described an enzymatic method for the simultaneous determination of ammonia and urea in seafood products. The method is specific and its results correlated well to most traditional spoilage tests.

| Day | Urea (mg/100g) | Ammonia (mg/100g) | mg TVN-N/100g | mg TMA-N/100g | Aerobic Plate Count |
|---|---|---|---|---|---|
| 1 | 1.41 | 16.53 | 17.54 | 3.22 | - |
| 3 | 1.48 | 11.68 | 13.28 | 2.77 | $9.8 \times 10^4$ |
| 5 | 0.55 | 10.12 | 12.95 | 4.03 | - |
| 7 | 0.19 | 4.42 | 11.25 | 3.81 | $3.6 \times 10^6$ |
| 9** | 0.93 | 9.04 | 13.56 | 2.68 | $1.9 \times 10^7$ |
| 11** | 1.52 | 9.83 | 16.56 | 5.19 | $4.8 \times 10^7$ |

Table 2. Ammonia, urea, total volatile nitrogen (TVN), trimethylamine (TMA) and aerobic plate count (APC) of flounder stored on ice.*

* All data were average of two samples. ** Samples stored in plastic bags.

| Day | Urea (mg/100g) | Ammonia (mg/100g) | mg TVN-N/100g | mg TMA-N/100g | Aerobic Plate Count |
|---|---|---|---|---|---|
| 1 | 70.30 | 41.73 | 43.85 | 4.79 | - |
| 3 | 92.60 | 49.66 | 51.18 | 3.62 | $2.8 \times 10^7$ |
| 5 | 36.70 | 72.39 | 74.68 | 6.80 | - |
| 7 | 113.20 | 123.47 | 98.94 | 12.98 | $1.9 \times 10^8$ |
| 9 | 119.22 | 132.72 | 115.36 | 13.60 | $2.7 \times 10^8$ |
| 11 | 147.67 | 185.72 | 119.73 | 15.21 | $3.9 \times 10^8$ |

Table 3. Ammonia, urea, total volatile nitrogen (TVN), trimethylamine (TMA) and aerobic plate count (APC) of crabmeat stored in plastic bags on ice.*

* All data were the average of two samples.

## REFERENCES


American Public Health Association. 1970. Recommended Methods for Microbiological Examination of Food. 2nd ed. APHA, Inc., New York.

Biochemical Information. 1980. Methods of Enzymatic Food Analysis, Boehringer Mannheim Biochemicals, Indianapolis, Indiana.

Burnett, J.L. 1965. J. Assoc. Off. Anal. Chem. 48: 624-627.

Chaney, A.L. and Marback, E.P. 1962. Chin. Chem. 8: 130-132.

Cobb, B.F. III, Alaniz, I. and Thompson, C.A. Jr. 1973. J. Food Sci. 38: 431-436.

Conway, E.J. and Cooke, R. 1939. Biochem J. 33: 457-470.

Fawaz, G., and Dahl, K.V. 1964. Chem. Abstr. 61: 2177.

Hillig, F., Shelton, L.R. Jr. and Loughrey, J.H. 1959. J. AOAC. 42: 702-708.

Iyengar, J.R., Visweswariah, K., Moorjani, M.N. and Bhatia, D.S. 1960. J. Fish. Res. Bd. Canada. 17: 475-485.

Kaplan, A. 1969. Methods of Biochemical Analysis. John Wiley & Sons, New York. 17: 311-324.

Knight, C.B. and Toom, P.M. 1980. Proc. Fifth Trop. Subtrop. Fish. Tech. Conf. of Amer. Sea Grant College Program, Texas A&M Univ., College Station, TX 5: 181-194.

Mondzac, A., Ehrlick, G.E and Seegmiller, J.E. 1965. J. Lab Clin. Med. 66: 526-531.

Seligson, D. and Hirahara, K. 1957. J. Lab. Clin. Med. 49: 962-974.

Stansby, M.E. 1976. Industrial Fishery Technology, 3rd ed. Krieger Publ. Co., Huntington, N.Y. p. 370.

Steinbrecher, K. 1973. J. Assoc. Off. Anal. Chem. 56: 598-601.

Talke, H. and Schubert, G.E. 1965. Klin Wochenschr. 43: 174-175.

Vyncke, W. 1978. J. Food Technol. 13: 37-44.

Ward, D.R., Finne, G. and Nickelson, R. II. 1979. J. Food Sci. 44: 1052-1057.

# DETECTION OF BONES IN WHITE FISH FILETS USING ELECTRONIC CANDLING

Lester F. Whitney and Siraj Officewala
Professor and Research Assistant
Food Engineering Department
University of Massachusetts
Amherst, MA 01003

INTRODUCTION. Fish sticks (fingers) and fish portions have until recently become increasingly popular since their introduction in the 1950's. Initially, fish sticks and fish portions were prepared from teleost or minced fish flesh. Frozen fish filet blocks are also used as the raw material in the manufacture of fish sticks and portions. Since the most suitable species such as haddock and cod-fish have diminished alarmingly from over fishing, alternative sources need to be considered using such underutilized species as pollock, whiting and hake.

There has been at least one commercial recognition of and interest in the trend. For example, one processor in Rhode Island mechanically fileted whiting for fish filets with a yield of less than 20% of the weight of a whole fish.. The processor would like to filet the fish in such a way that 25-30% of weight of the whole fish could be retained. However, a 5-10% increase in filet weight would lead to an increasing incidence of fish bones because the machine knives need to be adjusted closer to the skeleton. Adequate quality control requires either normal inspection or an automatic machine which can detect fish bones and separate the defective filets from acceptable "bone-free" ones, or automatically eject bones allowing acceptable filets to proceed to the next unit operation (Whitney, 1979).

The fish processing industry also labors under some confusion as to what is a bone and what is a "non-bone". The U. K. Association of Frozen Food Producers (Anonymous, 1978b) adopt the following definition of a "Fish-Bone":

a. Any bone which is less than 10 mm long and less than 3 mm in any other dimension would not be regarded as a bone for the purpose of being classified as a defect.

b. Any bone which is greater than 40 mm long and 10 mm in any other dimension would be regarded as a "Critical Bone".

c. Any bone which exceeds the 10 x 3 mm limit but is less than the 40 x 10 mm limit should rate as a "Bone Defect".

Because of such recommendations, many times a fish processor suffers while getting poorly graded for its products, with correspondingly lowered profit. In general, filets with no bones (or any dimension) are preferred by processors. In the fish industry, bone detection is usually done manually by visually inspecting each filet or using the sense of touch.

It is desirable that a dependable mechanical/electrical method for bone detection be developed for better quality control of filets. Fish bones and flesh (without striations) absorb light in different amounts. This is also true in the case of x-rays; but here detection is limited to the wavelength and intensity of light produced by a fluorescent screen. This requires a very high voltage power supply (Jaundrell-Thompson, 1970) with attendant dangers in an environment subject to water encroachment; also, high costs of invested equipment are involved. X-ray equipment has not been adopted by this industry for these and many other reasons.

A well-defined beam of light which impinges on a photomultiplier tube, produces a certain average DC voltage output dependent on the wavelength and intensity of that light. When such a beam of light as passed through a "white" fish filet is cut by a moving object such as a bone, an instantaneous drop in the average DC voltage is provided, dependent upon the amount of light absorbed. This instantaneous change in average DC voltage is seen as a relatively low voltage signal on an oscilloscope. Such a technique can be made practical for the detection of fish bones with the applications of appropriate electronics and instrumentation.

## OBJECTIVES

To effect an increase in the use of underutilized species, such as whiting, for fish products, one must overcome the problem of obtaining acceptable "non-bone" defective filets. Filet suppliers to processors must be able to identify bone-free filets, preferably by automatic means. While X-rays are well-known to detect bones inside the flesh of vertebrate animals, the method has not been adopted by the fishing industry for this purpose. Quite apart from this technological need, candling of eggs is a commercially available method to detect blood spots and/or oversize air cells within. When a "white" fish filet is held in contact with a light source, one can visually determine bones, if any exist, in much the same manner. In fact, candling tables are commercially available and in limited use in the fishing industry.

The objectives of this research were to investigate these methods as follows:

1. To experiment with X-rays and visible light sources for fish bone detection in filets.

2. To study the effect of candling fish filets electronically by measuring the voltage output of a photomultiplier tube produced from diffused light beam impinging upon fish bones with the following variables:

   a. two filet thicknesses, 6.35 and 12.70 mm
   b. two equivalent processing belt speeds, 0.1 and 0.2 m/s
   c. three equivalent belt speeds, for at least one bone of each of four* dimensions from one fish
   d. four different wavelengths as produced by colored "Polythane" films of red, blue, green and that from a bare incandescent light source; for at least one bone of each of four* dimensions from one fish.

## MATERIALS AND METHODS

Fresh haddock and codfish were selected and purchased from a local fish market because of availability. Carcasses of fresh haddock and codfish were dissected and selected bones were washed, air dried and stored individually in plastic boxes. Three bones were selected for each minimum dimension of 0.254, 0.381, 0.508 and 0.635 mm.

Filet portions were first prepared as 6.35 and 12.70 mm thick slabs, then cut to obtain specimen blocks. Formaldehyde was used to prevent fish spoilage while maintaining a constant filet sample to avoid variations from this source.

### Apparatus for Detecting Bone

A bone detection unit was fabricated as shown in Figs. 2 & 3. A plastic cup (internal diameter 24.8 mm, external diameter 51.7 mm and 23 mm high) held filet specimens up to 20 mm thickness. The plastic cup was completely lined and shielded with thin sheet metal (0.508 mm thick) except for an orifice to obtain a well defined beam of light through an active orifice diameter of 1.4 mm which can be made to impinge on the fiber optic below. The plastic cup could be raised or lowered along a rod standard. The plastic cup was 10.5 mm above the fiber optic to allow for the bone to pass freely through this space. Experiments proved that the position of the bone, either above, within or below the filet was of no consequence, and for reasons of experimental control, the bone specimens were passed under the fileted fish sample.

An alligator clip (7 mm high; 26.4 mm long) was soldered to a long screw (4mm; 76.6 mm long) which in turn was fastened radially into a rotating hub. The alligator clip held the fish bone in an extended position along the radial elevation. Fish bones were inserted into a sleeve of plastic wire insulation at the base end for better grip between the two jaws of the alligator clip to prevent crushing.

It should be noted that this apparatus for detecting bones must be kept enclosed in a dark chamber. The light transmitted by the fiber optic must only be from the incandescent light source.

*Four bone dimensions were arbitrarily selected to encompass previous work by other researchers.

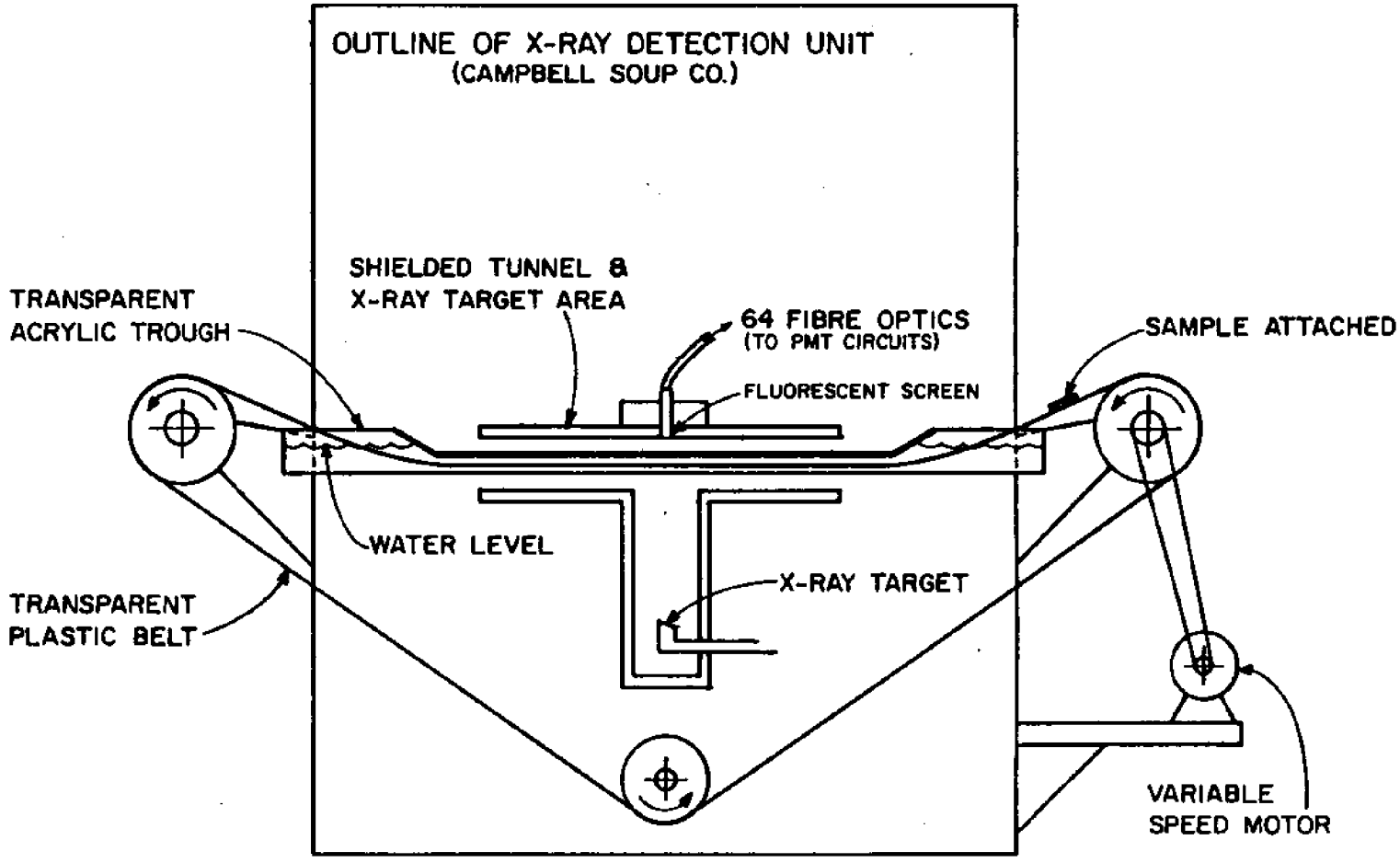


Fig. 1. Modified commercial bone detection X-ray unit supplied by Campbell Soup Co., Camden, New Jersey.

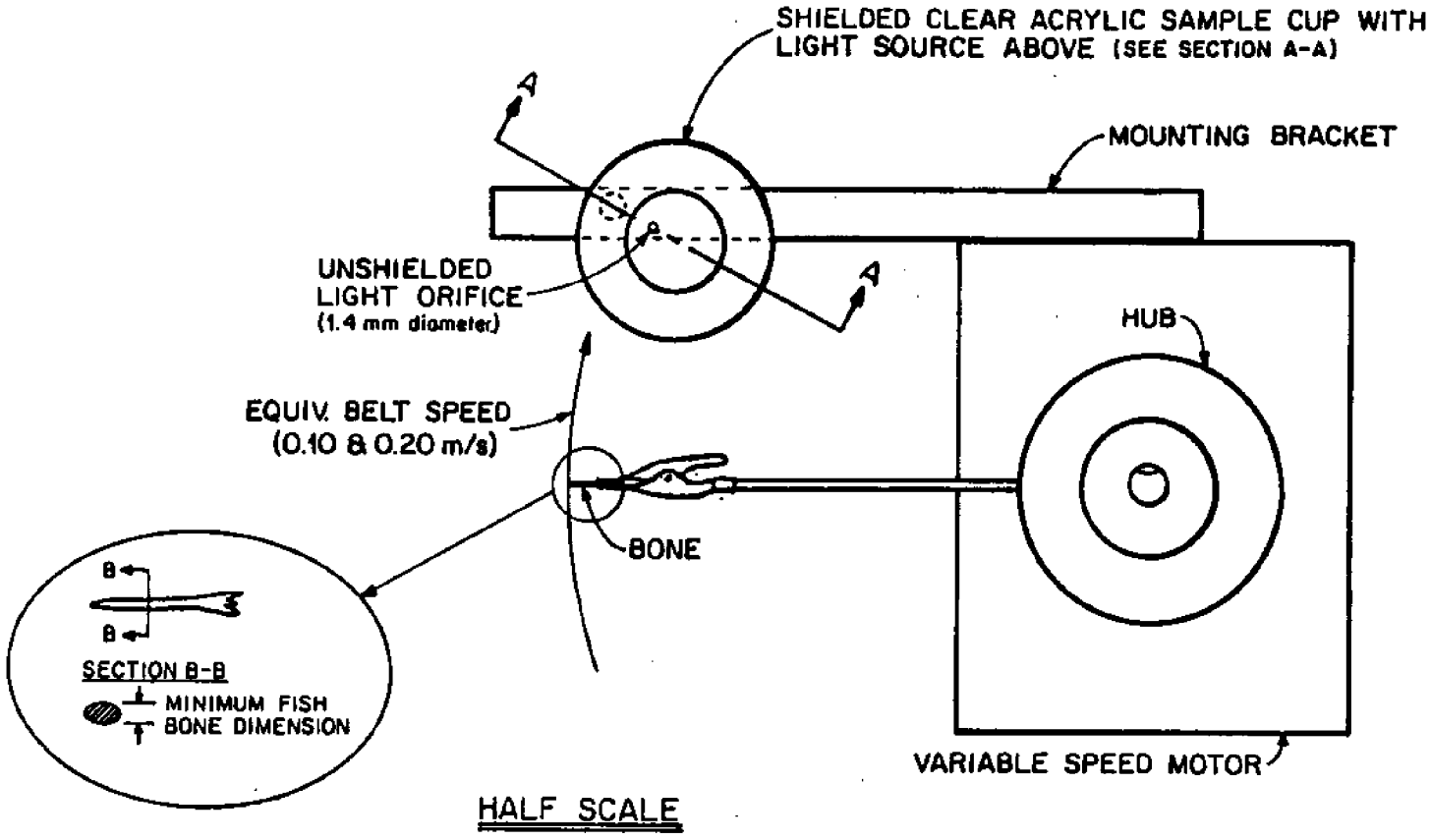


Fig. 2 . Schematic plan view of fish bone speed simulator under fish fillet sample with light candling apparatus.

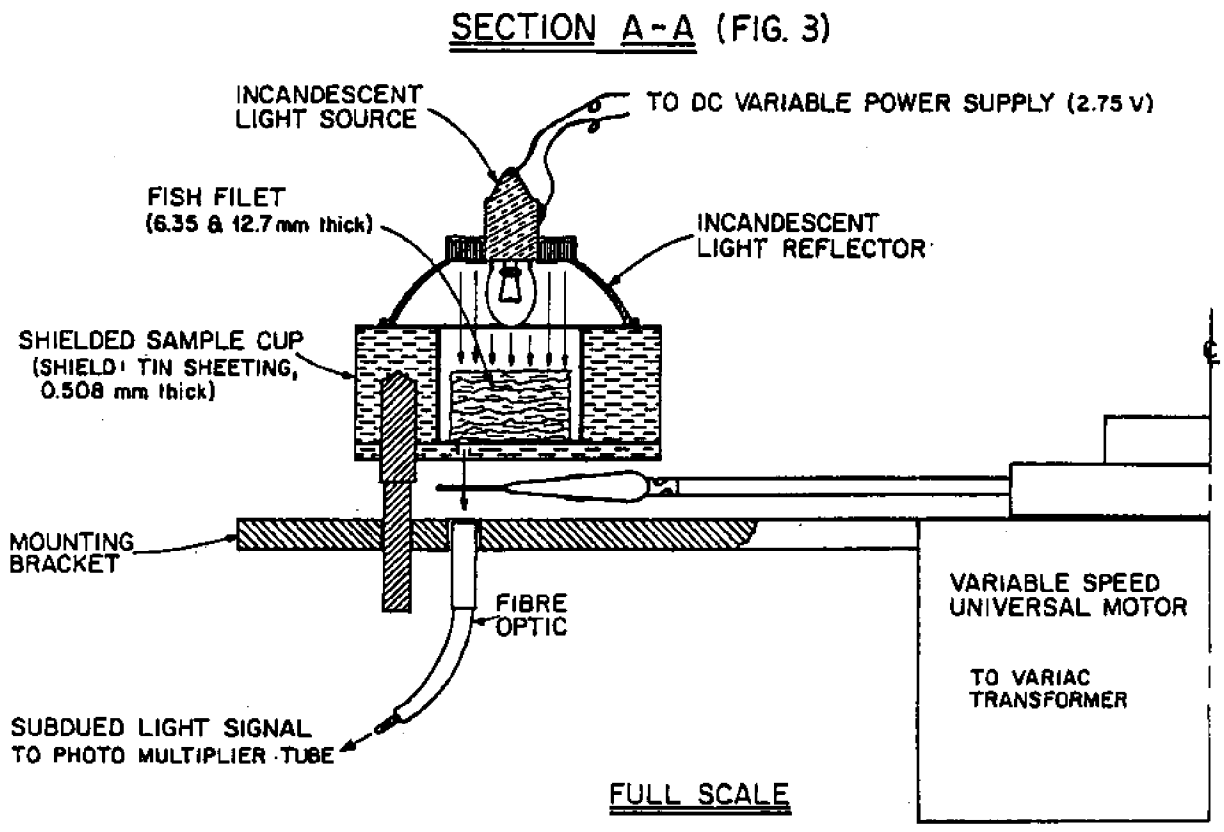


Fig. 3. Sectional elevation view of light candling apparatus through fish fillet and fiber optic pick up of intercepted signals.

Incandescent Light Source Power Supply

For the candling experiments, a common flashlight bulb and reflector was used as the incandescent light source. This was energized by a constant voltage power supply. The output level was adjusted to 2.75 volts at no load. The light source with its reflector was fixed over the stationary filet specimen holder as shown in Figures 2 & 3.

Turntable Motor

The electric motor selected was a universal gear motor manufactured by Buchler Instruments, single phase, 50-60 Hz, 120 vac. The motor could reliably produce revolutions from 7 rpm of radius arm 13.7 cms which corresponds to an equivalent belt speed of 0.1 m/s and up, at the center of the fiber optic sensor.

A variable autotransformer (mfgd. by SGA Scientific, Bloomfield, N.J.) provided a variable power supply to permit changes in velocity of the bone sample, as it passed over the fiber optic, with equivalent belt speeds of 0.1 and 0.2 m/s.

Oscilloscope

This oscilloscope (Type: R564B; Model: 121N; Serial: B020125; manufactured by Tektronix) had the facility for storage along with an auto-erase. In the left wing, a differential amplifier (Type: 2A63; Serial: 011415) measured voltages along the vertical scale. A time base (Type: 2B67; Serial 026113) provided in the right wing measured sweep time along the horizontal scale.

## RESULTS AND DISCUSSION

Each fish filet specimen (6.5 and 12.70 mm) from haddock and codfish species were placed in the specimen cup such that the thickness was measured along the light beam direction of travel as shown in Fig. 3.

There were two similar series of experiments conducted for haddock and codfish species. Each experiment was divided into four parts. For the first part, the speed of the hub was set at 15.2 rpm which corresponded to an equivalent belt speed of 0.2 m/s at the center of the fiber optic. The haddock filet specimen thickness was selected at 6.35 mm for the first part. Bones were oriented with their minimum dimensions measured along the light beam travel as shown in Fig. 2. For each of the size categories of bones (0.254, 0.381, 0.508 and 0.635 mm in turn at the point of interception of the fiber optic) three replicates were selected. Each bone was clamped in the alligator clip such that the corresponding minimum dimension passed over the fiber optics' active area. The motor was switched on to allow the bone to pass over the fiber optic at an equivalent belt speed of 0.2 m/s. Each time a bone passed over the fiber optic, the PMT produced an instantaneous drop in voltage output. Ten observations showing the maximum instantaneous drop in PMT voltage output was observed and recorded. All data were statistically

analyzed by computer for the interactions and inferences from the variables of this study. Typical results for the series of experiments are shown in Figures 4 and 5.

An attempt was also made to study the effect of an equivalent belt speed of 0.4 m/s on the drop in PMT voltage output. The results for one series of bone dimension at 0.1, 0.2 and 0.4 m/s and one filet thickness of haddock are presented in Fig. 6.

The photomultiplier tube responded differently when various wavelengths of monochromatic light were made to impinge on it. To study the nature of responses obtained from a PMT due to different wavelengths in the visible light spectrum, three colored "polythane" film filters (red, blue and green) were used as barriers over the bare incandescent light bulb. Only one thickness (12.70 mm) for haddock filet was used. The effects of one bone for each of the four different dimension categories were studied. The results are presented in Fig. 7. The ratio of voltage output signals for largest:smallest bone was also calculated, as shown in Fig. 8.

It should be noted herein that the selection of the technique for moving the bone under the filet specimen was a matter of providing the most controllable and reproducible experimental conditions. Voltage output from the PMT was determined for bone locations within the filet, as well as under and over the filet, with no discernible or appreciable differences. This single method facilitated rapid and easy changes in variable adjustments to permit constant, reproducible and measurable observations on variables for these experiments.

## SUMMARY AND CONCLUSIONS

A sufficient signal was produced to trigger a rejection system for all experiments; and these signals are of at least one order of magnitude greater than those reported for X-ray detection by similar electronic techniques. Thus, a generalized conclusion may be made that the method warrants further study as a means for automatically detecting bones in clear, white fish filets of commercial interest.

### Experiments With X-rays

1. X-rays are satisfactory for general purpose detection, and can be employed for any fish flesh, even with striation or skin in place, providing sufficient time is available for such detection.

2. Bone detection is limited to the wavelength and intensity of light produced by a fluorescent screen.

3. X-rays penetrate throughout all the unshielded area of the bone detection apparatus; the fluorescent screen transforms X-rays to light energy on its surface, but at a very low level of lumen output. This was insufficient to produce meaningful PMT voltage outputs as for small fish bones.

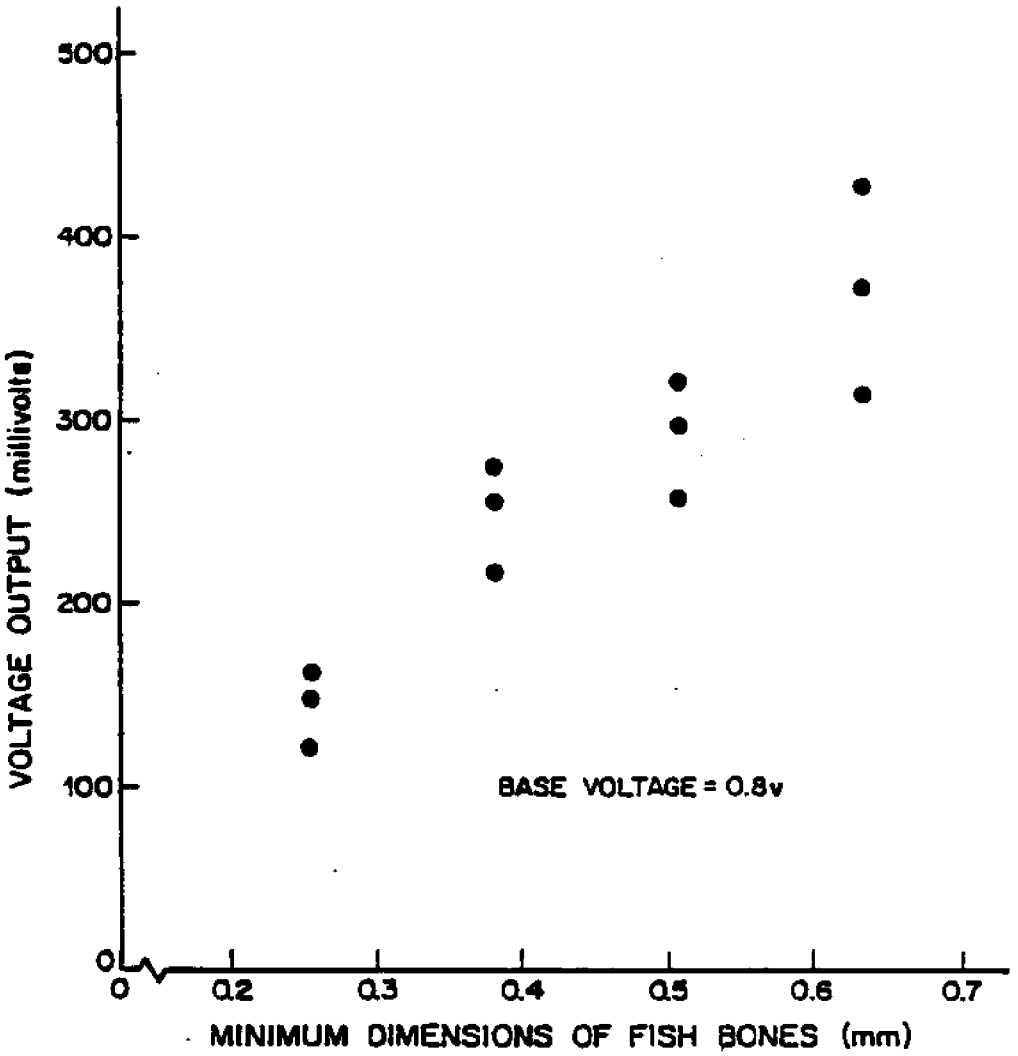


Fig. 4. Photomultiplier tube voltage output versus haddock bone size at 0.2 m/s and 12.70 mm fillet thickness.

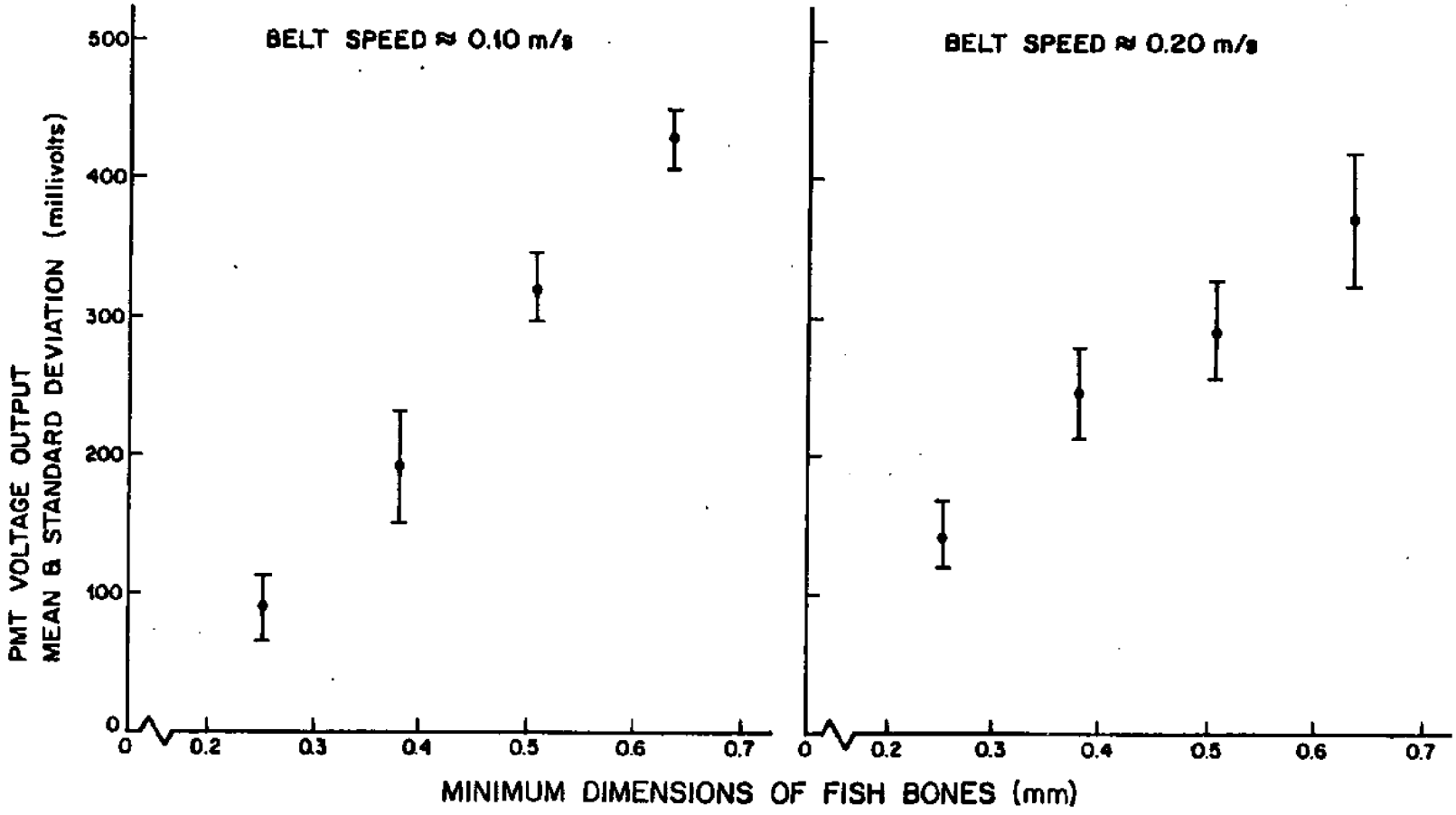


Fig. 5. Comparison of effects of two bone speeds on PMT voltage output for 12.70 mm haddock fillet thickness.

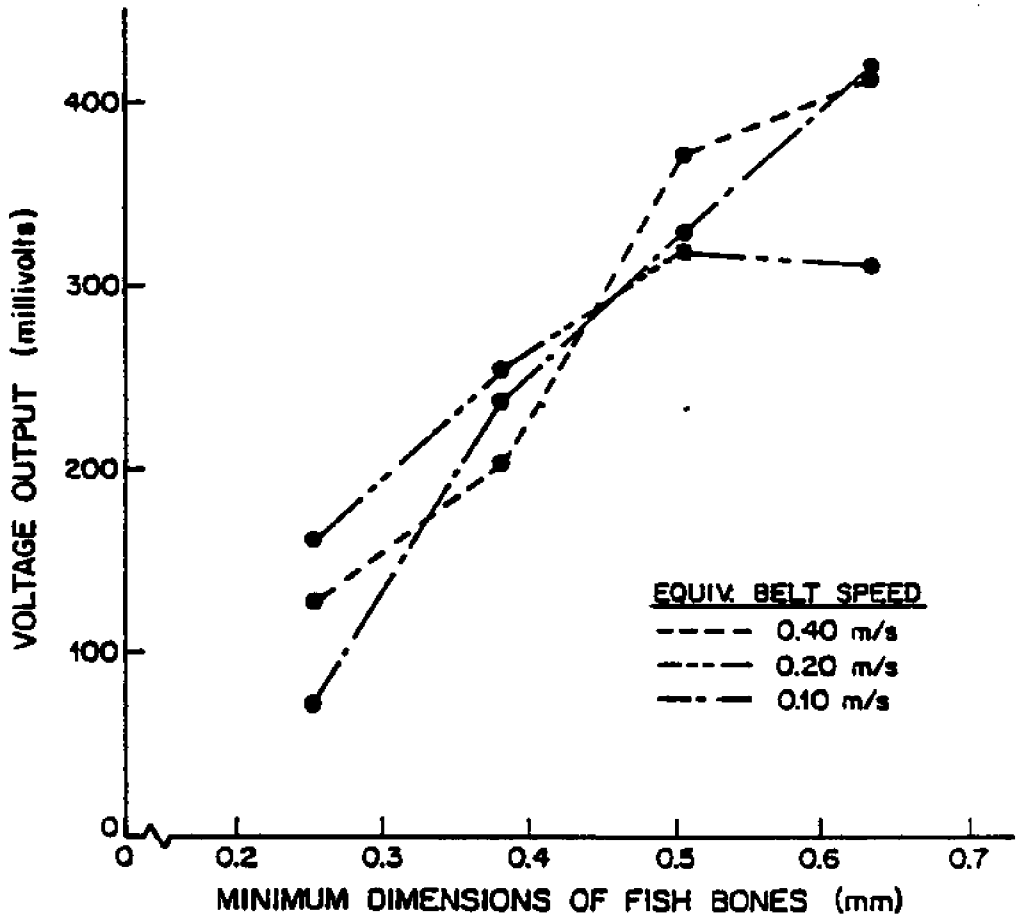


Fig. 6. Photomultiplier tube voltage output versus haddock bone size and 6.35 mm fillet thickness as affected by three speeds.

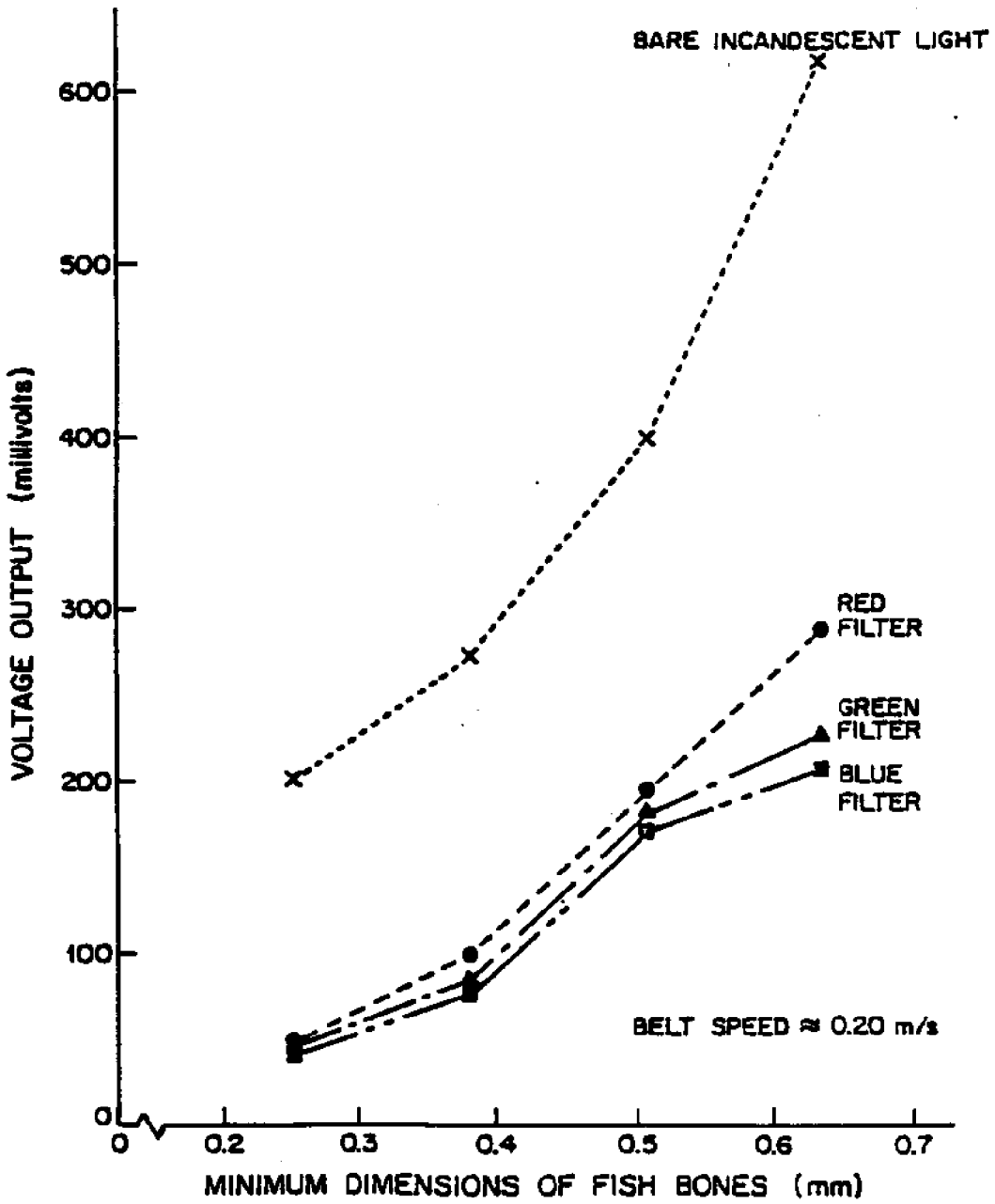


Fig. 7. Comparison of effects of incandescent light of different wavelengths on PMT voltage output.

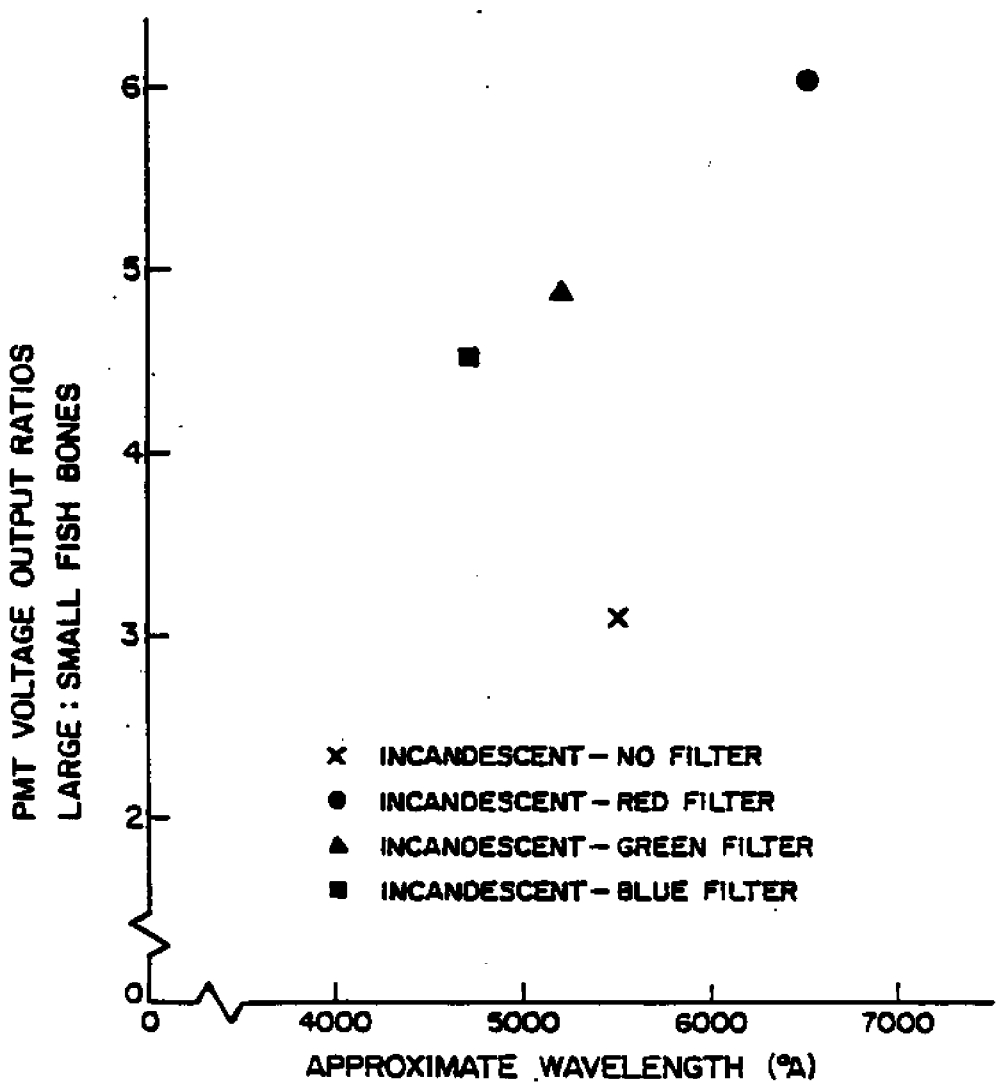


Fig. 8. Photomultiplier tube voltage output ratios at different wavelengths.

Candling With Light

4. Bare incandescent light can be controlled as a well defined beam, as compared to X-rays. Strong PMT output signals resulted from bones intercepting such subdued light beam passing through a filet.

5. Bare incandescent light was the most suitable source of energy for candling white fish filets (without striations) for bone detection as compared to filtered incandescent light.

6. The flesh characteristics of a fish species produces different results in output signal. For example, a 6.35 mm haddock filet with "flaky" flesh produces twice as much drop in the base level voltage output as a similar codfish filet. Thus a particular species will produce a particular set of data as input for controlling the bone detection operations.

7. Speed of interception of a light beam by a bone as studied was of no apparent consequence. Moran et al (1) made similar conclusions for their X-ray experiments.

8. Bones of the same minimum dimension produced highly significant results as PMT voltage outputs. This may be due to minor differences in bone density for the same minimum dimension. Barkley & Dexter Laboratories, Inc., made a similar conclusion during their preliminary X-scan testing for fish bone detection in conjunction with this work. They further suggested in their report that the limit of bone detection must be defined by bone density and not by bone diameter.

In conclusion, detection of fish bones in white fish filets without striation can be effectively accomplished with bare incandescent light as a candling method at industrial belt speeds of up to 0.4 m/s. This compares to typical processing belt speeds of 0.15 m/s.

REFERENCE

1. MORAN, J. M., WISE, D. P., TETRAULT, R. and CARVER, J. H. 1965. Prototype automatic fish-bone detector. Food Technol. 19(5):46.

Research supported by U.S. Department of Commerce, NOAA, N.E. Fisheries Development Program and Massachusetts Experiment Station, University of Massachusetts at Amherst.

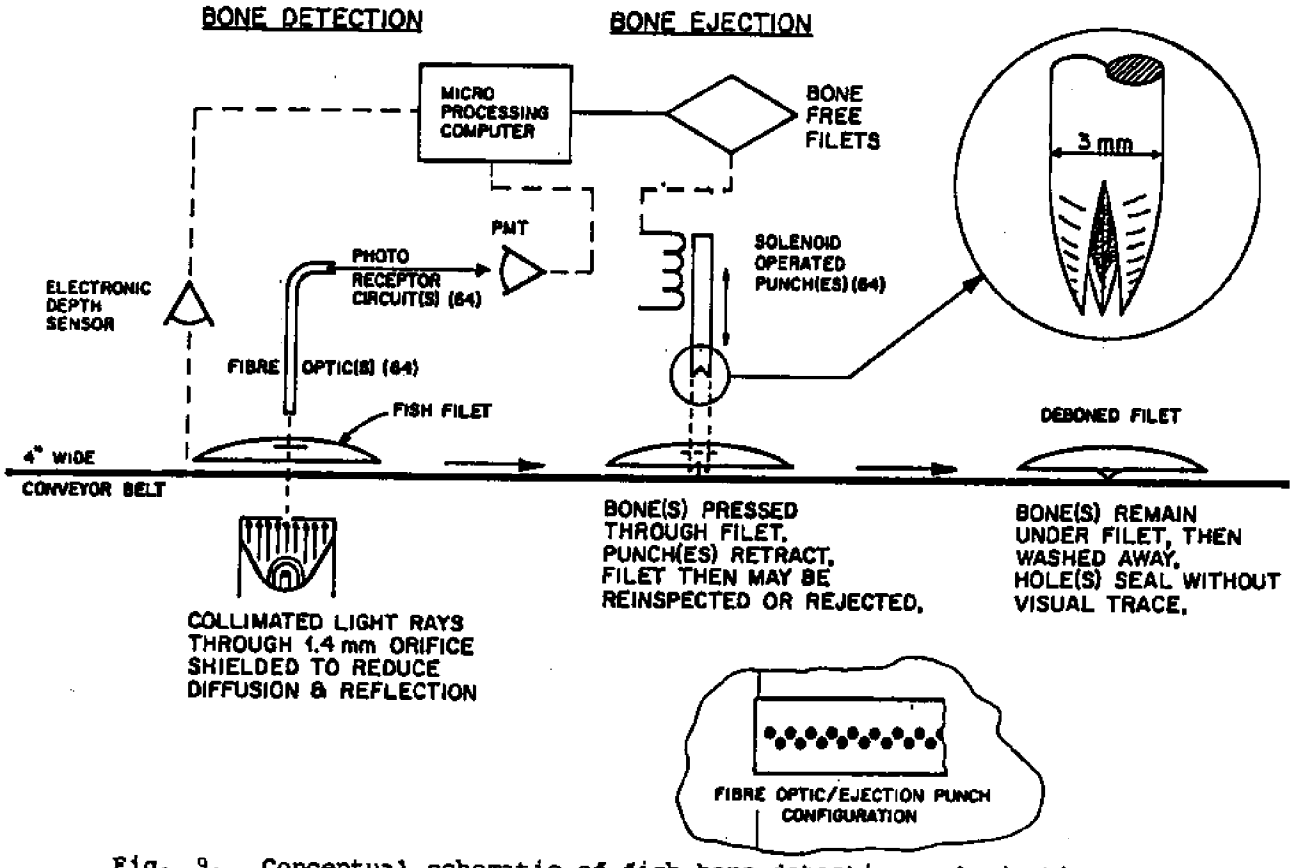


Fig. 9. Conceptual schematic of fish bone detection and ejection system using incandescent light candling.

# EXPLORATION OF MENHADEN AS A RESOURCE FOR SURIMI PRODUCTION AND USE IN SIMULATED SHELLFISH PRODUCTS

Tyre C. Lanier, Takayuki Akahane, and Reino Korhonen
Department of Food Science
N. C. State University
Raleigh, NC 27650

## INTRODUCTION

This report summarizes work conducted to develop processing procedures for production of high grade surimi from menhaden. Surimi is a bland, light colored, and stabilized wet (frozen) concentrate of fish muscle proteins which is a key functional component of many restructured seafood products now consumed in Japan and the United States, primarily due to its unique texture-forming (gelation) properties. Unlike the "fish protein concentrates" of two decades ago, surimi is made only from wholesome fish muscle, free of any bones, skin, head, or intestinal parts of the fish. It is prepared by mechanical separation of fish muscle from headed and gutted fish of prime quality, washing of the minced flesh with fresh cold water to remove blood and other undesirable soluble constituents, and dewatering by pressing to attain a moisture content similar to intact muscle. Cryoprotective agents such as sugar and sorbitol are normally added to protect the gel-forming properties of the fish proteins during frozen storage so that the surimi may be held eight months or more prior to usage in further processed food products.

## MATERIALS AND METHODS

Thus far, several pilot plant trials and one commercial trial have been completed. The pilot plant trials were conducted at the NCSU Seafood Laboratory in Morehead City, N.C. Fish were obtained directly from the water aboard a menhaden fishing vessel during normal fishing operations. The fish were held iced

until docking (within 4-6 hours) and immediately headed, gutted, and washed. Surimi was processed either the same day or the following morning. Fish were deboned on a Yanagyia "mini" model drum-type deboner with 3 mm orifices. Each ten lb. batch of deboned meat was mixed with 30 lb. of iced tap water for each of three successive washes and stirred for 5 min in a large Hobart mixer at low speed. Sodium bicarbonate (0.5%) was added to the first wash to stabilize the pH, while 0.15% salt (sodium chloride) was added to the final wash to facilitate dewatering. The slurry was dewatered between washes by pouring through a nylon mesh bag. The final dewatering was accomplished by centrifuging the mesh bag containing the mince for about 15 min in a Bock commercial centrifuge in a cold room. A strainer had not been obtained at the time these tests were run; therefore, the washed mince was directly chopped in a silent cutter with 4% sucrose and 4% sorbitol added to protect the proteins during storage. The surimi thus produced was wrapped in polyethylene bags and frozen at -20°C until determination of the gel-forming ability could be made.

Gels were prepared by chopping tempered surimi with 2% salt (NaCl) and added ice to adjust the moisture: protein ratio to 5:1. This batter was chopped in a special laboratory vacuum cutter to a temperature of +5°C and then transferred to a plastic bag. The bag was vacuum sealed to remove air pockets, a corner slit open, and placed in a sausage stuffer where the cold batter was extruded without air pockets into stainless steel tubes, 0.5 in I.D. by 6 in. long, sealed at both ends by a stopper and a screw-on brass cap. The tubes were processed by one of three schedules: (a) 40°C (104°F) for 1 hour, followed by a 10 min cook at 90°C (194°F). This process reveals the "setting ability of the protein at low temperatures and can reveal the presence of low temperature protein-degrading enzymes. (b) 60°C (140°F) for 30 minutes. This process will reveal the presence of so-called "alkaline protease" enzymes which can degrade proteins and deteriorate texture. (c) 90°C (194°F) for 10 min. This rapidly cooks the batter without the influence of low temperature setting or protease activity.

The tubes were cooled in ice after processing, the gels removed and stored in plastic bags under refrigeration before evaluation of the gel texture.

The texture of each gel was evaluated using a 2-bite compression test between two plates on an Instron Universal Testing Machine. One inch long

samples were subjected to a cross-head speed of 100 mm/min and compressed radially to 74% of their thickness. "Toughness" of the samples is defined as the maximum force attained on the first bite divided by the sample weight. "Cohesiveness" is defined as the ratio of peak force of the second bite to the peak force of the first bite, being an indication of how much structural integrity the sample retains after being compressed one time. Toughness and cohesiveness values have been directly related to the "gel strength" and folding test" values obtained by traditional Japanese methods in this laboratory, and can be made available for comparison if desired.

Subsequent to the laboratory/pilot plant trials, a commercial trial was arranged at the facilities of Nichibei Fisheries, Inc., Bayou LaBatre, Ala. Approximately 350 lbs (headed and gutted) of Atlantic menhaden were trucked in from Reedville, Va on ice, being approximately 2 1/2 days post-mortem at time of surimi processing. These fish had been headed and gutted, but not thoroughly washed (judging from the bloody appearance of the fish interior) within 6-8 hours following landing. Care was taken in the harvest and subsequent handling to rapidly attain and maintain a low temperature in the fish. Additionally, 2000 lbs of whole Gulf menhaden were harvested locally by a small day boat, immediately iced onboard and transported to the Nichibei plant. These fish were headed and gutted soon after delivery (6-8 hours from harvest). Two batches were processed that day, being differentiated by the addition of 0.5% sodium bicarbonate to the first wash of batch 1, with no bicarbonate used for batch 2. The following day, a third batch of the same fish was processed along with the Atlantic menhaden which arrived that morning. Both were processed with the addition of bicarbonate to the first wash. To all four batches 0.10% NaCl was added to the final (third) wash to facilitate dewatering in the screwpress.

## RESULTS

The results of several pilot trials conducted were similar such that the data were averaged for the North Carolina menhaden samples, as shown in Figure 1. Comparison is made with identically obtained properties of commercial samples of Gulf croaker surimi and Alaskan pollack surimi (grade SA-Japanese produced) as well as with surimi prepared experimentally in an identical manner from New England red hake. The experimentally-produced menhaden surimi demonstrated

excellent gelling properties at 90°C and with the 40°-90° treatment, which are commonly used for the manufacture of restructured shellfish meats. Surprisingly, however, the gelling properties at 60°C were extremely poor. The cohesiveness value for this gel is misleadingly high, as this type of measurement does not reflect true cohesiveness when a sample is extremely mushy in its texture.

It is obvious that a proteolytic enzyme(s) was present in the menhaden surimi which is extremely active near 60°C, but which has little activity at 40° or 90°C. This type of enzyme(s), termed alkaline protease, has been extensively studied by this laboratory. It is found in the mince of most mechanically deboned fish and commonly originates primarily from visceral organs, either by contamination due to improper cleaning of fish prior to deboning, or by leaching of the gut juices into the muscle tissue during iced storage of whole fish. However, menhaden used in this study were cleaned thoroughly and fairly soon after catching, yet the alkaline protease activity was much more intense than has been encountered for any other species studied to date, however poorly handled. If the thermal processing of restructured foods containing such surimi is precisely controlled and the product diameter/thickness is small, the presence of the protease should pose little problem. However, elimination of the protease would be much more desirable, as the surimi could be used without restriction in any restructured seafood product.

Several trials were conducted to improve the color of surimi prepared in the pilot plant, as samples prepared by conventional procedures (minus a strainer, which was not then available) were quite dark in appearance. Tests were made of fish deboned at various belt pressures on the deboner with careful alignment of butterflyed fillets in a meat-side-down position on the drum, in an attempt to separate the more loosely held light meat from the dark meat which lies next to the skin. This approach was not successful, however. Addition of bicarbonate to the first wash did seem to increase the extraction of blood and muscle pigments from the light flesh. At this time, further work to improve the color was postponed until more sophisticated separation techniques, similar to those developed by the Japanese for processing sardines into surimi, could be developed.

Yield data obtained on the four batches of the commercial trials at Nichibei (Table 1) do not reflect

Table 1. Approximate Yields of Commercial Trial at Nichibei Plant

| | % of Whole Fish |
|---|---|
| Heading, Gutting | 50-60 |
| Deboning | 27.5-33 |
| Washing, Straining | 11.5-14.0 |
| Straining Waste | 3.8-5.0 |

Table 2. Proximate Composition of Fish Processed at Nichibei Plant

| Description | Moisture | Protein | Fat | Ash |
|---|---|---|---|---|
| Gulf Menhaden | | | | |
| whole fish | 67.7 | 15.0 | 13.5 | 4.4 |
| deboned fish | 76.6 | 16.9 | 7.0 | 0.4 |
| surimi* | 78.4 | 16.3 | 5.2 | 0.2 |
| strainer waste | 75.0 | 17.3 | 6.9 | 0.8 |
| Atlantic Menhaden | | | | |
| H&G fish | 70.3 | 16.8 | 11.7 | 2.8 |
| deboned mince | 75.4 | 18.4 | 5.9 | 1.1 |
| surimi* | 80.1 | 18.1 | 2.0 | 0.2 |
| strainer waste | 75.4 | 20.1 | 3.9 | 0.8 |

*taken directly from strainer, no cryoprotectant addition.
values are averages of batches 1, 2, and 3.

maximum attainable yields. Adjustment of the processing machinery and larger batch runs (to minimize constant routine losses encountered in filling and running the machinery) should result in higher yields during normal commercial processing.

Table 2 contains compositional data for whole or H&G fish, mechanicIly separated mince, and surimi (before addition of cryoprotective additives). For both species, the process results in a product which has increased moisture content and increased protein content (the protein being composed primarily of functional salt-soluble proteins rather than less desirable water soluble proteins as in natural muscle), and a significantly reduced fat and ash content. The color of the surimi was very light and greatly improved over that of the deboned mince. Specks of dark meat were apparent in the waste taken from the strainer. This waste was higher in protein and lower in water content than the top-grade surimi, with a higher fat and ash content. The extra fat in the waste is likely associated with the bits of skin and dark meat which were removed, while the increased ash is likely due to small bone fragments removed by the strainer. A second pass of this material through the strainer would be expected to yield a good quality second-run product and would increase the overall yield of the process.

Table 3 presents a compositional analysis of the wash waters from both species. Use of the bicarbonate in the first wash resulted in a 0.5 to 2.0 unit rise in pH over the initial pH of the meat. This treatment should be helpful in preventing denaturation (loss of gel-forming ability) of proteins which may occur upon prolonged exposure to the lower muscle pH which accompanies the onset of rigor mortis. The dry solids content for the first wash water of the Gulf menhaden was higher than that of the Atlantic menhaden, as was evidenced by more suspended fibers and fragments. As might be expected from the higher initial fat content of the Gulf fish (Table 2), a much higher fat content was found in the wash waters of this species, particularly in the first wash. The actual amount of fat removed in the washes is not apparent from this data; samples were taken by opening a submerged container within the water after settling of the fish had occurred. The greater proportion of fat removed from the Gulf menhaden floated to the surface, such that it was not included in the sample. The greater turbidity and solids content of the Gulf menhaden first wash can be explained by the higher fat content of the dry material. This would reduce the specific gravity of

Table 3. Analysis of Wash Waters in Commercial Trial at Nichibei Plant

pH of Wash Waters

| Description | Batch No. | Before Washing | 1st Wash | 2nd Wash | 3rd Wash |
|---|---|---|---|---|---|
| Gulf Menhaden | 1 | 6.4 | 6.9 | 6.5 | 6.5 |
| Gulf Menhaden | 2 | 6.3 | * | 6.5 | 6.8 |
| Gulf Menhaden | 3 | 5.8 | 7.8 | 7.0 | 6.8 |
| Atlantic Menhaden | 4 | 6.2 | 7.4 | 7.2 | * |

*not measured

Appearance of Wash Waters

Gulf Menhaden

Waste water from first wash: The water was turbid and showed dark yellow color. It contained more total solid such as a small fragment of meat and fat tissue, and more fat which formed thin layer.

Waste water from second wash: The turbidity was lower than that of first waste water. The total solids decreased.

Waste water from third wash: The water was most transparent among these three waste waters. It showed a light yellow color.

Atlantic Menhaden

Waste water from first wash: The water showed a more bloody color. The small fiber of meat settled down. The fat content was lower than that of Gulf menhaden.

Waste water from second wash: The water showed a yellow color. The amount of small fibers of meat decreased. The fat content was lower than that of the first wash.

Waste water from third wash: The appearance was similar to the second waste water.

Table 3 (continued)

Composition of Wash Waters

| Sample name | Washing Solution | Dry solids contents (%) | Fat content (mg/g wash water) | % Fat of Dry Material |
|---|---|---|---|---|
| Gulf menhaden* | | | | |
| First wash | 0.1% $NaHCO_3$ | 1.22 | 4.03 | 33.0 |
| Second wash | Water | 0.69 | 0.83 | 12.0 |
| Third wash | 0.1% NaCl | 0.49 | 0.23 | 4.7 |
| Atlantic menhaden | | | | |
| First wash | 0.1% $NaHCO_3$ | 1.02 | 0.58 | 5.7 |
| Second wash | Water | 0.72 | 0.53 | 7.4 |
| Third wash | 0.1% NaCl | 0.67 | 0.47 | 7.0 |

*Samples were obtained on batch 3.

many particles causing them to float, and not settle as easily. Net salt soluble protein loss was calculated by subtracting total water soluble protein content from total salt soluble protein content. Losses of salt soluble (functional) protein were approximately the same for all three wash waters of the Gulf menhaden, but progressively decreased with each successive wash of the Atlantic menhaden.

Differences in appearance and composition between washes were greater for the pilot plant trials than the commercial trials. The design of the washing tanks used at Nichibei allowed for only half to two-thirds of the wash water to be skimmed off between washes; thus the washing process was not as thorough using such tanks as in the laboratory pilot plant where complete dewatering between washes is employed.

The bloody appearance of the first wash for Atlantic menhaden noted in Table 3 was likely the result of insufficient washing of the belly cavity of the fish during cleaning. The residual blood subsequently oxidized and gave a brownish appearance to the fish upon arrival in Alabama. The surimi made from these fish was also much more brown in color than the Gulf menhaden surimi. This emphasizes the need to thoroughly wash fish immediately after cleaning to remove residual blood.

The gelling properties of the four batches processed in the commercial trial are shown in Figure 1. The Gulf menhaden batches were all quite similar in gelling properties with the exception of the poor cohesive properties at the 90°C process of batch 2. Again, cohesiveness values of samples processed at 60°C are not truly indicative of the true cohesive properties of such samples, due to their mushy texture. The Atlantic menhaden surimi processed commercially (batch 4) was generally superior in gelling properties to the Gulf menhaden and greatly resembled the Atlantic menhaden surimi processed in the laboratory pilot plant. The superior gelling properties of the Atlantic menhaden may be due to one or more of several causes: (a) proteases in the Gulf menhaden may have some activity in the 40°C and 90° range; (b) the higher fat content of the Gulf menhaden gels may have adversely affected the gelling properties; or (c) the salt-soluble proteins of Gulf menhaden may be inferior in gelling properties to those of Atlantic menhaden. Future experimentation will be required to pinpoint the cause of the differences noted. The superiority of the commercially procesed Atlantic menhaden was quite

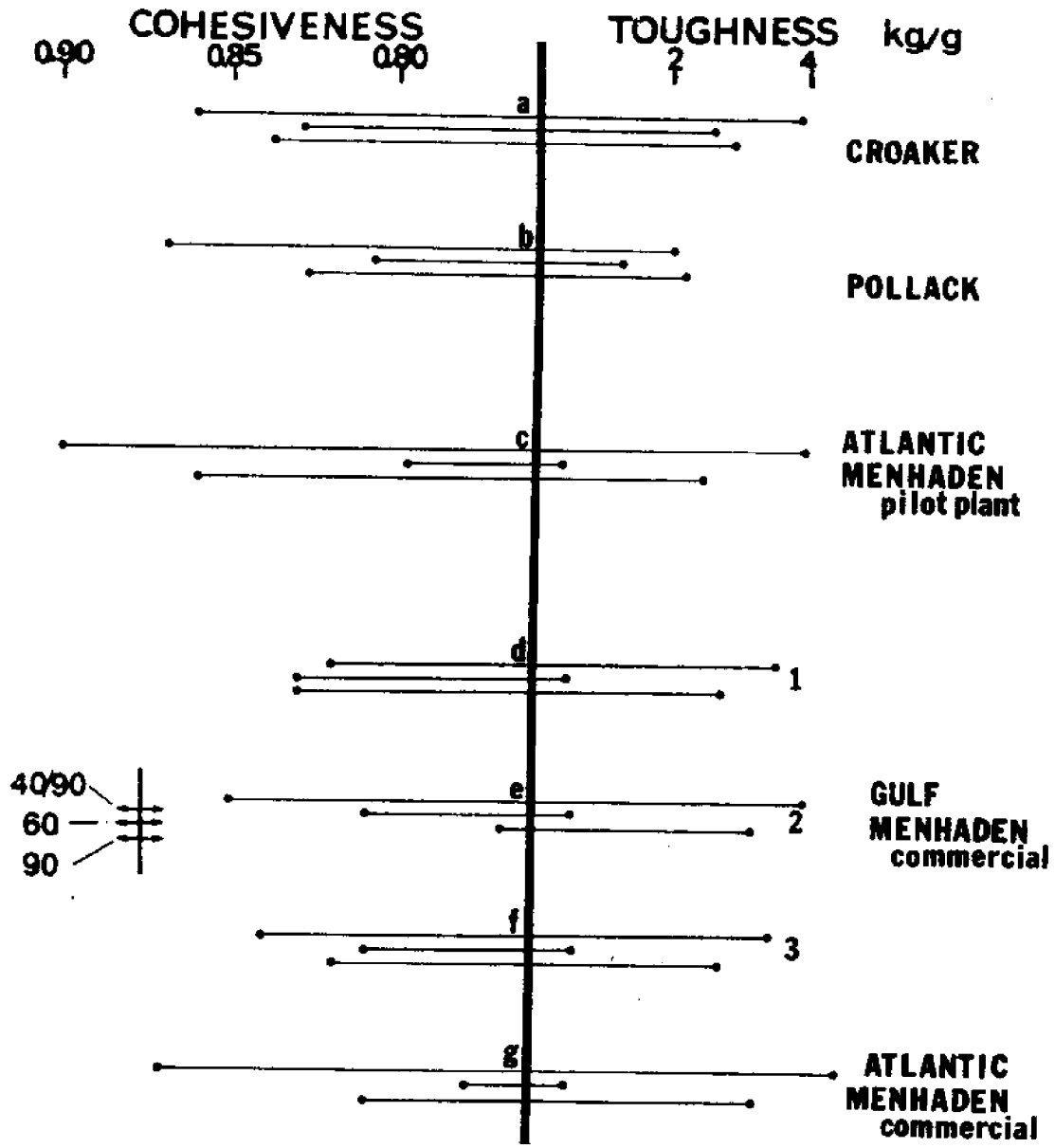


Figure 1. Comparison of textural values obtained on gels prepared from (a) commercial croaker and (b) pollack surimi with(c) Atlantic menhaden surimi prepared in the pilot plant, (d,e,f) three batches, 1, 2, and 3 respectively, of Gulf menhaden surimi produced in commercial trial, and (g) one batch of Atlantic menhaden produced in commercial trial.

surprising in light of its greater age post-mortem and generally poorer condition than the Gulf menhaden at time of processing.

The effectiveness of adding sodium bicarbonate to the first wash in terms of improved color or gelling properties is difficult to assess from the present data. Repetitive studies under more controlled conditions will be required to demonstrate whether or not the treatment has a definite effect.

## Summary

The following is a summary of the findings to date:

(1) Atlantic menhaden. Surimi had excellent gelling properties when processed at temperatures commonly used for restructured seafoods. However, the presence of a potent protein-degrading enzyme, active near 60°C, in samples processed thus far if not eliminated could preclude its use in products of large diameter or thickness (>0.5 inch). Efforts are currently in progress to eliminate this enzyme from the surimi, with a good likelihood of success. No samples have been processed thus far from this fish which have the lightness of color obtained with Gulf menhaden; however, no freshly processed and well cleaned fish have been used with the strainer as yet. The fat content of this surimi is quite low (2%). The stabilities of the fat and proteins during frozen storage of the surimi are currently being evaluated.

(2) Gulf menhaden. Surimi had excellent light to white color, a higher fat content (5%) and gelling properties somewhat less than Atlantic menhaden, yet nonetheless adequate for use in restructured products. Further work is needed to determine whether the gelling properties at all processing temperatures may be further improved by removal of the protease and reduction of the fat content. The fish evaluated in this commercial trial were not as high in fat content as may be encountered in the Gulf (13% versus up to 20% fat or more), yet a good yield of fat could be obtained by skimming the wash waters. Equipment has been developed in Japan for fat removal and recovery during processing of fatty fish into surimi. The fat recovered would be food-grade by FDA standards, and might have differences in composition from rendered fat which could result in additional industrial or food uses. Future work is planned to recover this oil, develop methods for optimum yield, and characterize the lipid content so that its commercial value may be assessed.

The outlook at this point for development of a high quality surimi from menhaden seems extremely bright. While the presence of a potent protease is a problem at present, it is highly doubtful that such a potent enzyme originates in the muscle tissue rather than being of gut origin. Experiments now in progress should answer this question soon. What is likely is that some on-board pre-processing of fish may be necessary to produce a protease-free product in the shore-based surimi plant. Stability of the fat and protein during frozen storage of the surimi is still a question at the present time, but again, on-going research should reveal this soon. Should stability pose a problem, several treatments are envisioned which likely will overcome the problem.

A remaining question is the allowable tolerance of fat in surimi at the levels which might result from processing of menhaden surimi by conventional means. This will be evaluated in the coming months. Better removal of fat from the washed mince may be possible by use of defatting equipment similar to that developed by the Japanese.

A prototype counter-current washing machine has been designed and will be constructed within 1-2 months. Meanwhile, some initial studies using small batches in sequence to simulate a countercurrent system will be used to evaluate the concept and develop some initial parameters for running the finished equipment. Such a system would be expected to increase the water-use efficiency of the surimi processing operation and possibly yield a more concentrated high-protein effluent for recovery and use as animal feed.

The present extensive market for menhaden by-products is certainly an added incentive to develop a suitable surimi product from this fish. Close proximity of a surimi plant could result in optimized and full utilization of this enormous resource. The added value would be tremendously beneficial, as would be the more direct contribution of menhaden to the food supply of the world.

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# UTILIZATION OF OSMOREGULATION IN PENAEID SHRIMP TO ENHANCE FLAVOR CHARACTERISTICS

V. Z. McCoid and G. Finne
Seafood Technology
Texas A & M University
College Station, Texas 77843

## INTRODUCTION

Consumption of fish and shellfish in the United States has been steadily increasing while commercial landings have remained relatively stable. In 1981, United States shrimp landings totalled 218.9 million pounds with a value of 463.4 million dollars. Imports during the same period totalled 222.7 million pounds valued at 723.8 million dollars, comprising 51% of total shrimp poundage and 61% of the total revenue (15). Increased demand comes at a time when the wild harvest is at or near maximum sustainable yield (6). Overfishing and environmental quality are factors which limit the commercial fishery and thus threaten the seafood industry (13).

In recent years extensive progress has been made towards the domestication of aquatic animals of a number of different species. Shrimp mariculture could play a major role in supplementing the supply of shrimp while providing a much needed new economic resource. Success of a mariculture industry may well depend on the quality of the food product. One important consideration in production of a quality product is flavor. Since free amino acids have been shown to function as osmoregulators in crustaceans (1, 8, 9, 16) and to be major contributors to the flavor of seafood (4, 5, 7, 11, 14), changes in environmental salinity were used in an attempt to produce shrimp with optimum flavor profiles.

## MATERIALS AND METHODS

I. Effect of environmental salinity on free amino acid concentration in shrimp.

Penaeus vannamei were obtained from the Texas A & M Mariculture facility in Flour Bluff. Mean weight of the shrimp was 12.43 grams with a mean length of 15.38 centimeters. Live animals were transported to the seafood technology laboratory and ten shrimp were placed into each of six fifty gallon aquaria of 35 parts per thousand (ppt) salinity and temperature of 28° C. Shrimp were then allowed to acclimate for 72 hours.

Following acclimation, salinity in each of the tanks was changed to one of the following: 10, 20, 30, 40, 50 or 60 ppt. Changes were made at a rate of 2 ppt/hour, 10 ppt/day using appropriate volumes of either fresh water or a concentrated salt solution. Two replicates were completed in this manner. Immediately before changing the salinity, one shrimp was removed from each tank to obtain initial values. During the first run, one shrimp was removed from each tank at 24 hour intervals for a period of eight days. During the second, samples were taken at 24 hour intervals over four days. Immediately after removal from the tank shrimp were deheaded, deveined and peeled. Extracts were prepared by blending shrimp in a Vir Tis "23" homogenizer at a ratio of 1 g shrimp to 2 ml of 7% trichloroacetic acid (TCA). The extract was centrifuged and the supernatant analyzed for amino acid nitrogen (AAN) using a modification of the copper procedure of Spies and Chambers (12) as described by Cobb et al. (2). Following this analysis the supernatant was frozen at -25° C for amino acid profiles performed on an automated Beckman model 120 C amino acid analyzer. After examination of AAN values, samples of 72 hours and 144 hours were chosen from each salinity for these profiles.

II. Rate of change in free amino acids with changing salinity.

Penaeus setiferus obtained from the Texas A & M Mariculture facility in Flour Bluff were transported live to the Seafood Technology laboratory in Corpus Christi. Mean weight of the shrimp was 10.69 grams with a mean length of 9.07 centimeters. Twenty-five shrimp were placed into each of six fifty gallon aquaria of 35 ppt salinity and temperature of 28° C and allowed to acclimate for 72 hours. Following acclimation, three of the aquaria were changed to 10 ppt salinity and three were changed to 60 ppt salinity. These changes were made all at once with appropriate volumes of either fresh water or a concentrated salt solution. Immediately prior to changing the salinity, one shrimp was removed from each tank to obtain initial values. One shrimp was then taken from each tank at four hour intervals for a period of 28 hours and examined for AAN as described above. Samples were then frozen at -25° C for later use in amino acid profiles.

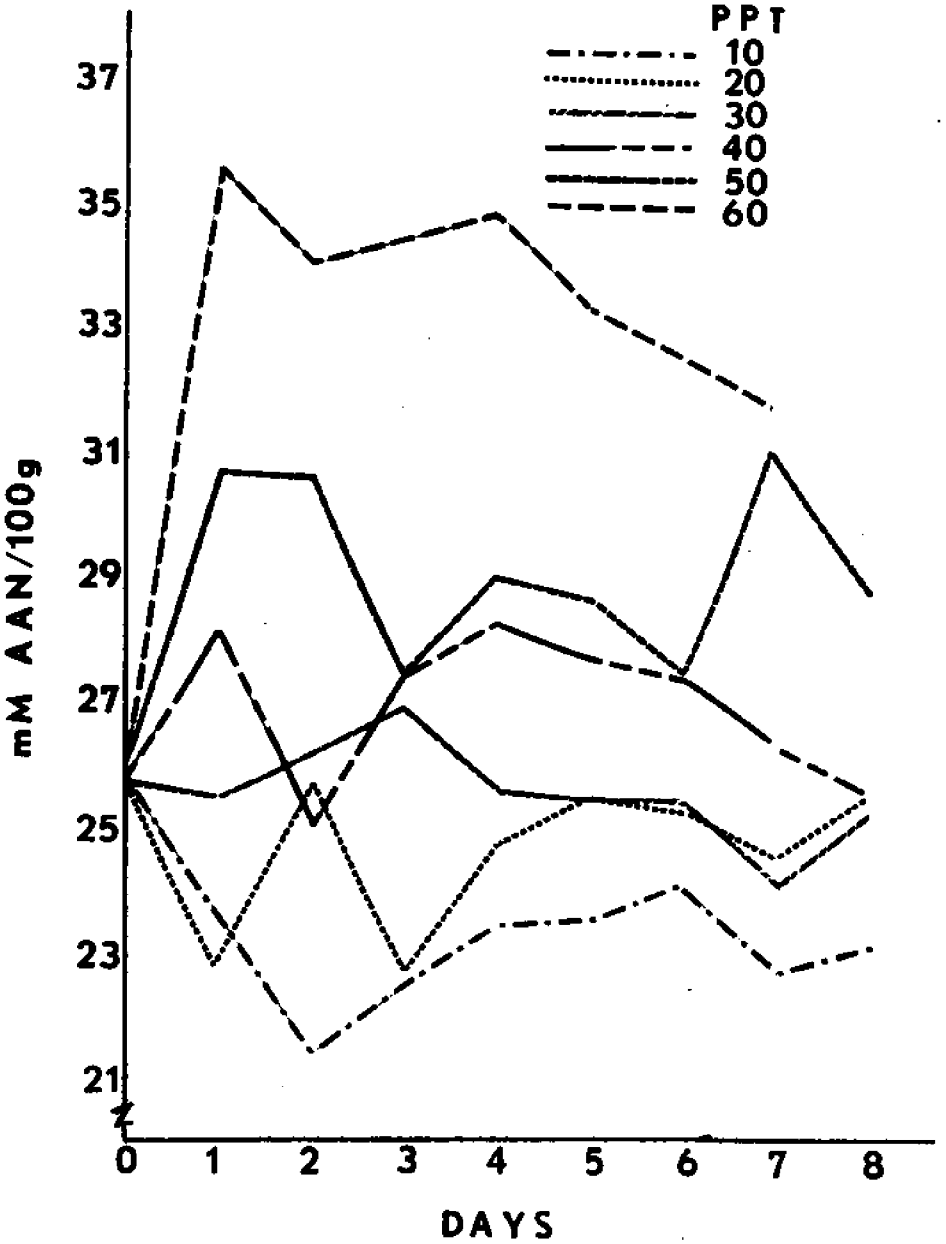
After determining that 49.03% and 54.26% of the maximum change in AAN at 10 ppt and 60 ppt, respectively, occurred within four hours the rate experiment was repeated. Penaeus setiferus with mean weight of 10.82 grams and mean length of 9.55 centimeters were used. Acclimation and salinity changes were done as described above. Shrimp samples were removed at 15 minute intervals for the first hour, 30 minute intervals for the next 3 hours, and at 8 hours, 24 hours and 48 hours. These samples were analyzed for AAN as previously described.

## RESULTS AND DISCUSSION

Figure 1 shows the response of Penaeus vannamei to changes in salinity over an eight day period. The free amino acid nitrogen was 25.77% mM/100g shrimp on day 0, an average of the values obtained from all six of the aquaria at 35 ppt. The maximum change in free amino acid concentration occurred within twenty-four hours with the exception of the samples taken from 10 ppt and 30 ppt which reached maximum change in 48 and 168 hours, respectively. (Values for the first four days represent the average from both replications.) Maximum changes in free amino acid concentration were -4.37 mM/100g, -2.97 mM/100g, -1.77 mM/100g, 2.33 mM/100g, 5.67 mM/100g, and 9.73 mM/100g for 10, 20, 30, 40, 50 and 60 ppt, respectively. The free amino acid concentration appeared to be relatively stable after 72 hours. Figure 2 shows a linear regression on salinity vs. AAN ($\hat{Y}$ = 20.504473 + .19221636) on values from 72 hours and following. The dotted line is through the means of the actual AAN values obtained with the black bars representing the range of these data points. These findings agree with other researchers (3, 8, 9, 10, 16) in demonstrating that free amino acid concentration in marine invertebrates varies with external salinity.

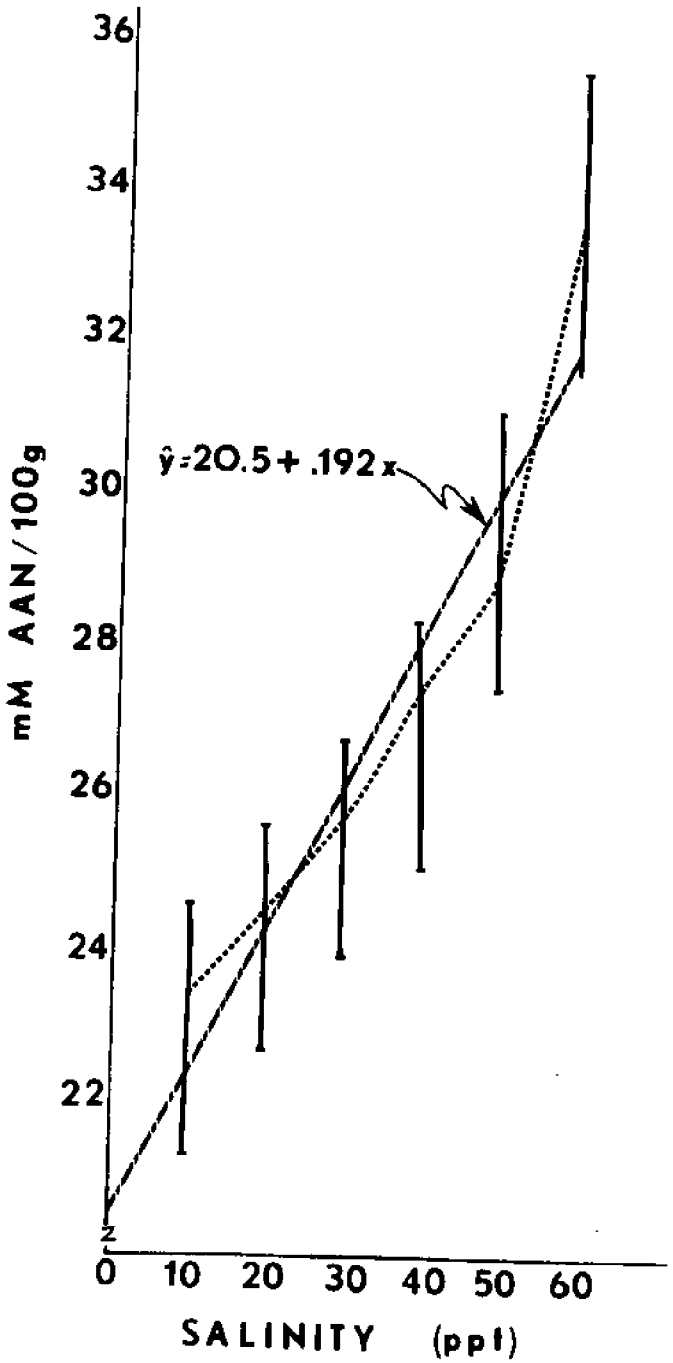
Tables 1 through 7 show the amino acid profiles represented as both molar percent and weight percent of sixteen amino acids. Values for threonine and serine are listed together since the process used could not separate these two amino acids efficiently. From these tables it can be observed that the molar percent of each amino acid fluctuates only slightly within and among the samples from the various salinities. These variations are probably attributable to experimental error since no apparent pattern is present. In all cases, glycine, proline, arginine, threonine/serine and alanine are present in the highest concentrations, together comprising 93% to 96% of total amino acids.

Percent change in total free amino acids is shown graphically in Figure 3. The zero % line represents the initial free amino acid concentration of samples taken at 35 ppt. Maximum percent change is as follows: 10 ppt = -16.95%, 20 ppt = -11.52%, 30 ppt = -6.87%, 40 ppt = 9.04%, 50 ppt = 22.00% and 60 ppt = 37.76%. Changes which occurred in the shrimp from decreased salinities were not as extreme as those from increased salinities. Figure 4 depicts the molar % of the amino acids present in highest concentrations. While the proportion of alanine appears to increase as salinity increases, glycine, arginine, proline and threonine/serine comprise an equivalent proportion regardless of salinity. Schoffeniels (9) shows that during regulation of intracellular osmotic pressure in the Chinese crab the concentrations of arginine, alanine, aspartic acid, glutamic acid, glycine and proline exhibit the most notable changes. This does not, however, appear to be true for P. vannamei. Although total free amino acid concentration does vary with environmental salinity, relative concentrations of the individual amino acids are fairly constant.




Millimoles of Amino Acid Nitrogen of Shrimp Maintained at Six Different Salinities

Figure 1




Millimoles of Amino Acid Nitrogen in Shrimp as a Function of Salinity

Figure 2

Table 1

Amino Acid Profiles

Shrimp on Day 0 at 35%.
25.20 mM AAN/100g

| Amino Acid | Molar % | Weight % |
|---|---|---|
| LYS | .86 | 1.27 |
| HIS | .45 | .72 |
| ARG | 13.79 | 24.95 |
| ASP | .10 | .13 |
| TAU | 1.09 | 1.35 |
| THR/SER | 6.23 | 6.79 |
| GLU | .98 | 1.46 |
| PRO | 21.33 | 23.99 |
| GLY | 46.13 | 30.48 |
| ALA | 6.57 | 5.41 |
| VAL | .54 | .62 |
| MET | .35 | .54 |
| ILE | .33 | .43 |
| LEU | .81 | 1.06 |
| TYR | .27 | .51 |
| PHE | .17 | .29 |

Table 2

Amino Acid Profiles

Shrimp Maintained at 10‰

| Amino Acid | 72 Hours 21.76 mM AAN/100g Molar % | Weight % | 144 Hours 24.00 mM AAN/100g Molar % | Weight % |
|---|---|---|---|---|
| LYS | 1.00 | 1.44 | .79 | 1.09 |
| HIS | .24 | .37 | .43 | .58 |
| ARG | 17.47 | 30.69 | 18.93 | 32.06 |
| ASP | .29 | .37 | .11 | .61 |
| TAU | 1.59 | 1.87 | .81 | .94 |
| THR/SER | 4.32 | 4.58 | 7.21 | 7.18 |
| GLU | 1.34 | 1.91 | 1.37 | 1.92 |
| PRO | 21.92 | 24.13 | 20.78 | 21.89 |
| GLY | 46.72 | 29.97 | 43.86 | 27.14 |
| ALA | 2.85 | 2.28 | 2.43 | 1.73 |
| VAL | 1.02 | .50 | .90 | .96 |
| MET | .29 | .49 | .53 | .75 |
| ILE | .23 | .30 | .50 | .62 |
| LEU | .54 | .69 | .98 | 1.19 |
| TYR | .17 | .30 | .26 | .51 |
| PHE | .01 | .11 | .11 | .18 |

Table 3

Amino Acid Profiles

Shrimp Maintained at 20‰

| Amino Acid | 72 Hours 23.40 mM AAN/100g Molar % | Weight % | 144 Hours 25.20 mM AAN/100g Molar % | Weight % |
|---|---|---|---|---|
| LYS | .34 | .53 | .40 | .57 |
| HIS | .29 | .48 | .42 | .72 |
| ARG | 11.12 | 21.02 | 18.68 | 31.99 |
| ASP | .12 | .18 | .14 | .18 |
| TAU | 1.41 | 1.82 | 1.16 | 1.36 |
| THR/SER | 5.17 | 5.90 | 6.48 | 6.74 |
| GLU | .56 | .89 | .84 | 1.20 |
| PRO | 24.88 | 29.24 | 23.85 | 25.39 |
| GLY | 53.38 | 36.85 | 44.44 | 27.80 |
| ALA | 1.67 | 1.46 | 1.13 | .88 |
| VAL | .09[a] | .13[a] | .72 | .77 |
| MET | .30 | .48 | .40 | .58 |
| ILE | .07 | .10 | .30 | .36 |
| LEU | .41 | .56 | .57 | .70 |
| TYR | .13 | .26 | .31 | .53 |
| PHE | .06 | .10 | .16 | .23 |

[a] Amino acid hand integrated

Table 4

Amino Acid Profiles

Shrimp Maintained at 30‰

| Amino Acid | 72 Hours 26.80 mM AAN/100g Molar % | Weight % | 144 Hours 25.40 mM AAN/100g Molar % | Weight % |
|---|---|---|---|---|
| LYS | .42 | .62 | .60 | .86 |
| HIS | .43 | .67 | .39 | .61 |
| ARG | 14.91 | 26.78 | 17.32 | 30.37 |
| ASP | .08 | .11 | .12 | .16 |
| TAU | 1.19 | 1.47 | .91 | 1.10 |
| THR/SER | 6.07 | 6.58 | 4.82 | 5.11 |
| GLU | 1.16 | 1.73 | .86 | 1.24 |
| PRO | 22.92 | 25.60 | 24.71 | 26.94 |
| GLY | 48.91 | 32.09 | 46.64 | 29.88 |
| ALA | 1.50 | 1.22 | 2.17 | 1.73 |
| VAL | .66 | .76 | .39 | .44 |
| MET | .38 | .56 | .33 | .48 |
| ILE | .31 | .39 | .11 | .17 |
| LEU | .76 | .99 | .36 | .46 |
| TYR | .21 | .37 | .21 | .35 |
| PHE | .09 | .06 | .06 | .10 |

Table 5

Amino Acid Profiles

Shrimp Maintained at 40‰

| Amino Acid | 72 Hours 27.40 mM AAN/100g Molar % | 72 Hours 27.40 mM AAN/100g Weight % | 144 Hours 27.40 mM AAN/100g Molar % | 144 Hours 27.40 mM AAN/100g Weight % |
|---|---|---|---|---|
| LYS | .43 | .67 | .74 | 1.15 |
| HIS | .33 | .54 | .30 | .50 |
| ARG | 15.96 | 29.76 | 13.78 | 22.94 |
| ASP | .27 | .36 | .54 | .75 |
| TAU | 1.24 | 1.59 | 1.10 | 1.44 |
| THR/SER | 5.12 | 5.75 | 6.42 | 7.36 |
| GLU | .44 | .68 | 1.13 | 1.78 |
| PRO | 23.84 | 27.63 | 17.63 | 20.81 |
| GLY | 43.62 | 24.53 | 49.33 | 34.23 |
| ALA | 6.88 | 5.83 | 6.85 | 5.92 |
| VAL | .56 | .66 | .66 | .77 |
| MET | .28 | .43 | .41 | .66 |
| ILE | .19 | .26 | .22 | .30 |
| LEU | .52 | .71 | .60 | .83 |
| TYR | .21 | .40 | .20 | .39 |
| PHE | .11 | .20 | .09 | .16 |

Table 6

Amino Acid Profiles

Shrimp Maintained at 50‰

| Amino Acid | 72 Hours 27.40 mM AAN/100g Molar % | Weight % | 144 Hours 27.40 mM AAN/100g Molar % | Weight % |
|---|---|---|---|---|
| LYS | .50 | .79 | .32 | .51 |
| HIS | .27 | .44 | .12 | .20 |
| ARG | 13.01 | 24.12 | 13.06 | 25.28 |
| ASP | .40 | .55 | .37 | .53 |
| TAU | 1.05 | 1.34 | 1.00 | 1.32 |
| THR/SER | 5.52 | 6.18 | 2.82 | 3.29 |
| GLU | 1.27 | 1.94 | .71 | 1.14 |
| PRO | 19.84 | 22.84 | 17.22 | 20.72 |
| GLY | 48.12 | 32.56 | 57.84 | 40.86 |
| ALA | 8.86 | 7.47 | 5.85 | 5.15 |
| VAL | .14[a] | .19[a] | .22 | .27 |
| MET | .22 | .35 | .14 | .23 |
| ILE | .16 | .22 | .06 | .09 |
| LEU | .38 | .51 | .21 | .29 |
| TYR | .19 | .38 | .06 | .12 |
| PHE | .07 | .12 | — [b] | — [b] |

[a] Amino acid hand integrated

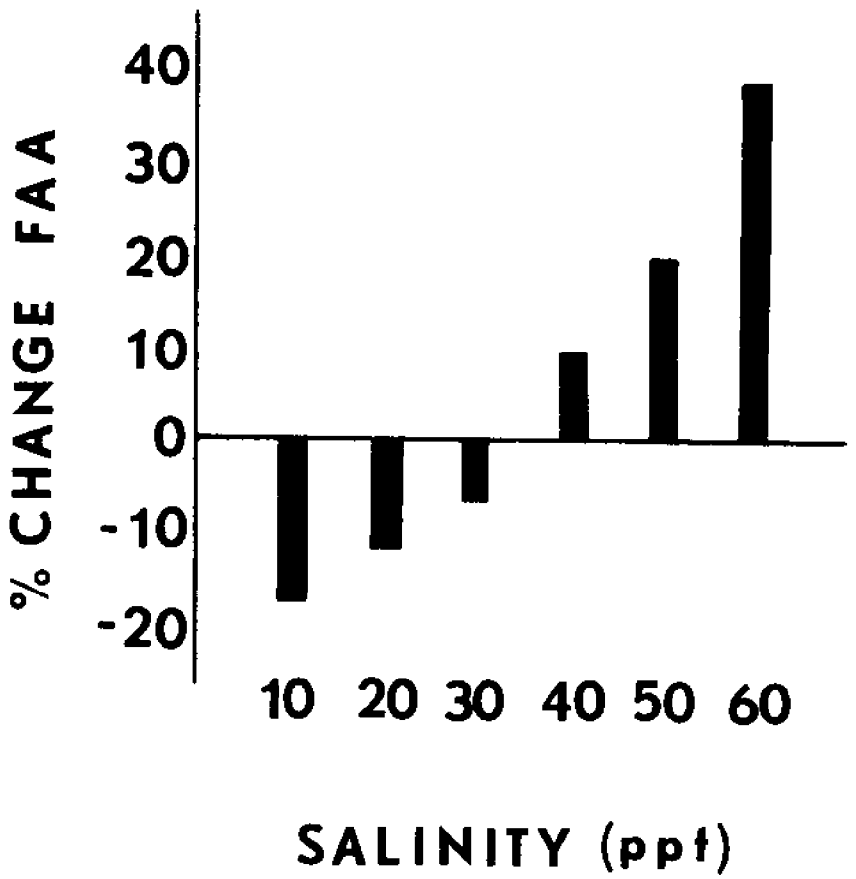
[b] Amino acid concentration too low for calculation

Table 7

Amino Acid Profiles

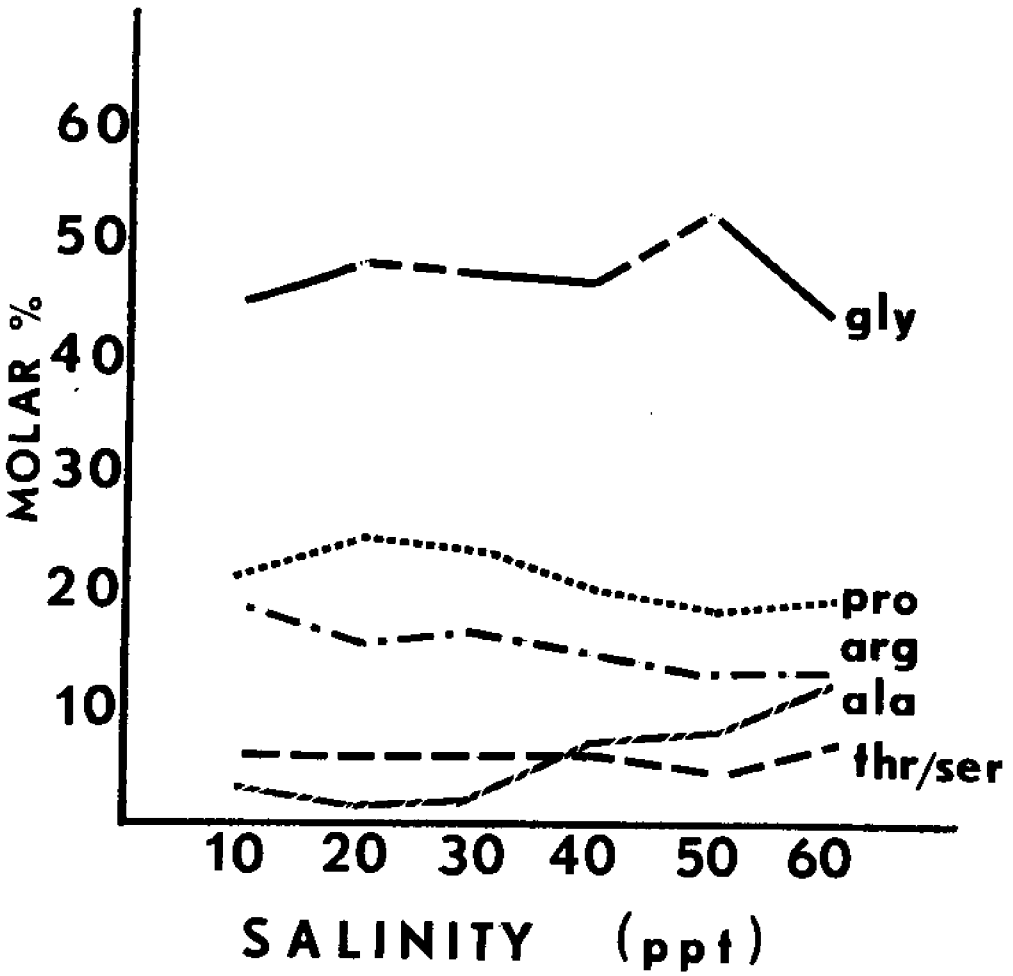
Shrimp Maintained at 60%.

| Amino Acid | 72 Hours 34.40 mM AAN/100g Molar % | Weight % | 144 Hours 32.48 mM AAN/100g Molar % | Weight % |
|---|---|---|---|---|
| LYS | .45 | .67 | .24 | .37 |
| HIS | .42 | .66 | .21 | .34 |
| ARG | 14.23 | 25.51 | 11.52 | 21.86 |
| ASP | .98 | 1.29 | .83 | 1.12 |
| TAU | .88 | .75 | .77 | 1.01 |
| THR/SER | 6.96 | 7.54 | 4.77 | 5.46 |
| GLU | .91 | 1.34 | .76 | 1.20 |
| PRO | 17.95 | 20.56 | 19.52 | 23.02 |
| GLY | 42.28 | 27.70 | 47.80 | 33.10 |
| ALA | 11.78 | 9.61 | 12.44 | 10.76 |
| VAL | .82 | .94 | .04 | .05 |
| MET | .65 | .98 | .30 | .48 |
| ILE | .43 | .56 | .16 | .22 |
| LEU | .74 | .95 | .40 | .55 |
| TYR | .35 | .66 | .17 | .34 |
| PHE | .17 | .28 | .07 | .12 |




Percent Change in Free Amino Acids (AAN) vs Salinity

Figure 3




Molar Percent of the Major Free
Amino acids at Various Salinities

Figure 4

Figure 5 shows the rate of change in free amino acid concentration for Penaeus setiferus. 49.03% and 54.26% of the maximum change at 10 ppt and 60 ppt, respectively, occurs within four hours of the salinity change. The low concentration of 20.89 mM AAN/100g and the high concentration of 33.72 mM/100g occur at 20 hours. Since such a large percentage of the change in free amino acid concentration occurred so rapidly, the experiment was repeated with analyses performed at shorter time intervals during the first four hours following the changes in salinity. These results are shown in Figure 6. Changes can be seen within the first 30 minutes. In 2.5 hours 49.03% of the maximum total change at 60 ppt occurs. During this same interval shrimp at 10 ppt have completed 82.00% of the maximum decrease in free amino acid concentration. This is followed by a slight rise and leveling off at three hours, with a downward trend resuming at eight hours.

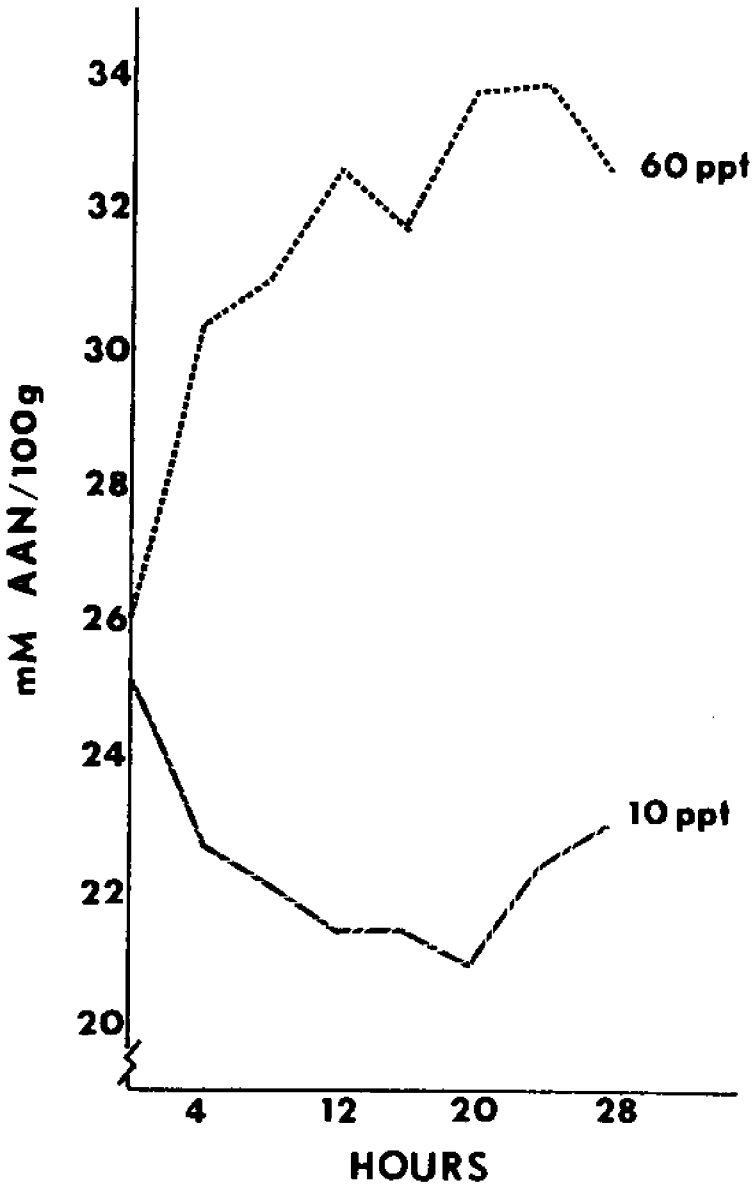
During the rate phase of this experiment adverse physical changes in P. setiferus were noted, which did not occur when the salinity changes were done gradually as in the first part of this research. In shrimp maintained at 60 ppt salinity, the muscle was softer and exuded water when peeled. This became apparent within 12 hours. The shrimp had an appearance similar to that of shrimp infected with microsporidia, commonly known as "cotton" shrimp. This change in appearance is a common, generalized reaction to stress situations in penaeid shrimp (A. L. Lawrence, personal communication). Nearly all of the affected shrimp had resumed their normal appearance within three days. The shrimp at both 10 ppt and 60 ppt also became less active during the first 24 hours following the rapid change in salinity.

## CONCLUSION

Both individual and mixtures of free amino acids exhibit inherent flavor characteristics (5, 14), with glycine, alanine, glutamic acid and methionine being the most influential (4). Although this research does not demonstrate large variations in molar percentage of any particular amino acid with respect to external salinity, there is a significant increase in total free amino acid concentration and, therefore, a molar increase in the individual constituents. In addition, maximum free amino acid values are achieved with 24 hours of salinity changes. Together these factors indicate that it is possible to produce a more flavorful penaeid shrimp, through mariculture, by utilizing manipulations of the environmental salinity.

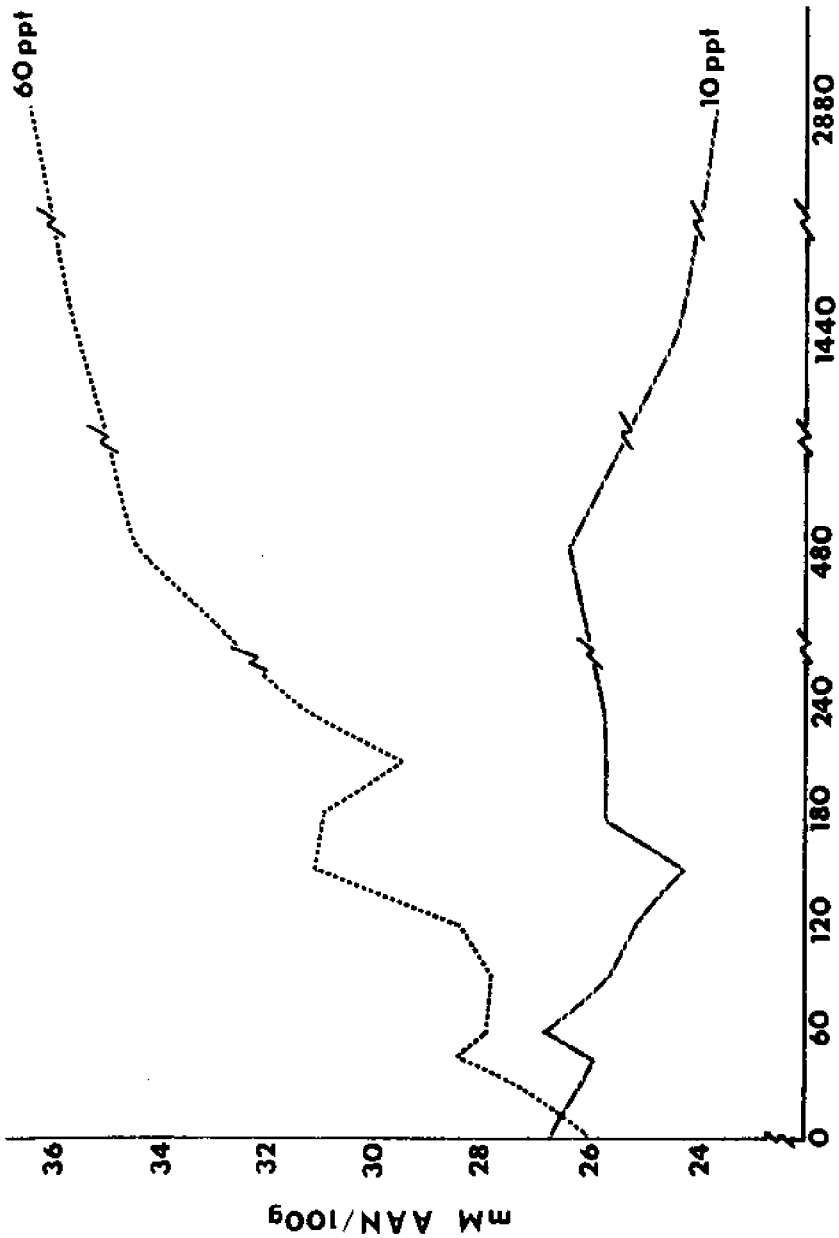
## REFERENCES

1. BEDFORD, J. J. and J. P. LEADER. 1977. The composition of the haemolymph and muscle tissue of the shore crab, Hemigrapsus edwardsi, exposed to different salinities. Comp. Biochem. Physiol. 57A:341-345.




Rate of Change in Free Amino
Acid Concentration (AAN)

Figure 5




Rate of Change in Free Amino Acid Concentration (AAN)

Figure 6

2. COBB, F. B. III, I. ALANIZ, and A. THOMPSON. 1973. Biochemical and microbial studies on shrimp: volatile nitrogen and amino nitrogen analysis. J. Food Sci. 38:431-436.

3. COBB, B. F. III, F. S. CONTE, and M. A. EDWARDS. 1975. Free amino acids and osmoregulation in penaeid shrimp. J. Agric. Food Chem. 23(6):1172-1174.

4. HASHIMOTO, Y. 1965. Taste-producing substances in marine products. p. 57-60. In: The Technology of Fish Utilization. Fishing News Books, Ltd., London, England.

5. JONES, N. R. 1969. Meat and fish flavors. Significance of ribonucleotides and their metabolites. J. Agric. Food Chem. 17:712-716.

6. LAWRENCE, A. L., G. W. CHAMBERLAIN, and D. L. HUTCHINS. 1981. Shrimp Mariculture. TAMU-SG-503. Texas A & M University Sea Grant College Program.

7. NAIR, M. R. and A. N. BOSE. 1965. Studies on post-mortem biochemical changes in prawns. p. 68-70. In: The Technology of Fish Utilization. Fishing News Books, Ltd., London, England.

8. ROSEIJADI, G., J. W. ANDERSON, and C. S. GIAM. 1976. Osmoregulation of the grass shrimp Palaemonetes pugio exposed to polychlorinated biphenyls (PCBs). II. Effect on free amino acids of muscle tissue. Mar. Biol. 38:357-363.

9. SCHOFFENIELS, E. 1967. Osmoregulation and nitrogen metabolism. p. 157-185. In: Cellular Aspects of Membrane Permeability. Pergamon Press, London, New York, Oxford, Toronto, Sydney, Paris, Braunschweig.

10. SCHOFFENIELS, E. and R. GILLES. 1970. Osmoregulation in aquatic arthropods. p. 255-286. In: Chemical Zoology. Vol. V, part A. Academic Press, New York and London.

11. SIMUDU, W. and M. HUJITA. 1954. Studies on muscle of aquatic animals - XXI. On glycine content in extractives of shrimps, with special reference to their taste. Bull. Jap. Soc. Sci. Fish. 20(8):720-721.

12. SPIES, J. R. and D. C. CHAMBERS. 1951. Spectrophotometric analysis of amino acids and peptides with their copper salts. J. Biol. Chem. 191:787-797.

13. STICKNEY, R. R. and J. T. DAVIS. 1981. Aquaculture in Texas: a status report and development plan. TAMU-SG-81-119. Sea Grant College Program Texas A & M University.

14. THOMPSON, A. B., A. S. McGILL, J. MURRAY, R. HARDY, and P. R. HOWGATE. 1979. The analysis of range of non-volatile constituents of cooked haddock (Gadus aeglefinus) and the influence of these on flavor. p. 484-487. In: Advances in Fisheries Science and Technology Fishing Books, Ltd., Farnham, Surrey, England.

15. THOMPSON, B. G. 1982. Fisheries of the United States, 1981. Bull. 8200. National Oceanic and Atmospheric Administration.

16. VINCENT-MARIQUE, C. and R. GILLES. 1970. Changes in amino acid concentration in blood and muscle of Eriocheir sinensis during hypoosmotic stress. p. 509-512. In: Life Sciences. Vol. 9, part I. Pergamon Press, Great Britain.

# CONSUMER PERCEPTION OF SULCASCARIS SULCATA IN SCALLOP ADDUCTOR MUSCLE

W. Steven Otwell and Bridget L. Walker
Department of Food Science and Human Nutrition
University of Florida
Gainesville, Florida 32611

## INTRODUCTION

Recent regulatory concern for parasitic nematodes, Sulcascaris sulcata in the adductor muscle of calico scallops, Argopectin gibbus has culminated in a 20 percent temporary action level (basis: one percent equals one nematode cyst per one adductor muscle per 100 muscle samples) specified by the U.S. Food and Drug Administration (FDA). Presence of this parasite and similar forms has been documented since 1930 (2) and described from the commercial regions for calico scallops in natural concentrations exceeding 38 percent infestation (3). Recent research to examine nematode infestation in calico scallops taken from 16 preselected stations between Savannah, Georgia and Fort Pierce, Florida in June, 1982 indicated all samples exceeded the 20 percent FDA limit (1). Regulatory inspections for nematode infestation have ranged in results from 0 percent to in excess of 45 percent[1]. These results provide speculation that there could be 'clean' areas (noninfested scallop beds) or 'less infested' scallop sizes or seasons.

Fortunately, FDA work referenced in publications (3, 4) and seminars[1] indicate the nematodes (S. sulcata) in calico scallops do not survive exposure to pH and temperatures which would normally be encountered during processing, cooking and human digestion. Thus, regulatory objection is essentially for aesthetic reasons. The objective of this work was to determine consumer perception of nematodes in calico scallops as a measure of the aesthetic problem.

## METHODS

All calico scallop samples were collected from processed and frozen meats (adductor muscle) which had been removed from commercial channels by an FDA directed voluntary action limit. Previous inspections declared over 40 percent of these scallop meats contained nematodes. The samples were obtained through a signed agreement

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[1]Florida Department of Agriculture and Consumer Services. 1982. State Regulatory Concerns, a discussion presented during the Industry/Regulatory Forum on Quality in Calico Scallop Production, organized by the University of Florida's Cooperative Extension and Sea Grant Advisory Services, December 2, 1982, Cocoa, Florida.

with FDA and the respective processing firm. Samples remained in frozen storage (-20°C) prior to tests. Thus, these samples were actual representation of adulterated product as determined by FDA.

Thawed samples were sorted by visual surface inspection to provide an infested and a noninfested group. These groups were later recombined to provide varying levels of infestation (0, 10, 20, 30, 40, 50, 75 and 100 percent infestation). Using a paired comparison test two levels of infestation were presented for consumer examinations. One level was always 0 percent infestation and the second level varied from 0 to 100 percent infestation, thus allowing eight pairs for comparable examination (pairs: 0 and 0 percent, 0 and 10 percent, 0 and 20 percent, etc... and 0 and 100 percent).

Each pair was displayed in two separate 9 inch aluminum pie pans resting side-by-side on crushed ice in a styro foam box. One pair was displayed per day. Volunteer consumers were asked to examine for differences between the two pans. Consumers were allowed to visually inspect, smell and touch the samples. They were not allowed to taste or consume samples. There was no limit on examination time per consumer. Days and time for displays were scheduled to insure adequate daylight.

Each display continued until responses were recorded for at least 100 consumers per displayed pair (table 1). All displays were conducted on the University of Florida campus in Gainesville, Florida. Consumers were prescreened to record consumer age and to assure an equal sex ratio per pair examined. There was no prescreening relative to the consumers opinion or previous consumption level for seafoods or scallops. The intent was to measure perception by the average consumer. No advise was given to direct consumer response. The consumers only knew they were examining scallops. All sensory responses were recorded. All accumulative responses per paired comparisons were calculated as a percent responding to a particular difference for the total consumers responding. Any reference to dots, spots, blotches, blemishes, specks, or 'those things' was recorded as a positive perception of nematodes.

One series of displayed pairs was conducted for raw scallops and a second series of displayed pairs was conducted for scallops pre-boiled at 250°C for 8 minutes with no addition of sauces, coverings, spices or water. The cooked series did not include paired comparisons with 75 and 100 percent infestation. The consumer was advised if the scallops were cooked.

## RESULTS AND DISCUSSION

These results represent the most stringent conditions in that the test design encouraged suspicion and careful examination. Consumers asked to examine for differences seemed obligated to detect differences. This reaction was obvious for responses which denoted differences that did not actually exist (table 2). For

Table 1. Characterization and total number of volunteer consumers participating in the tests.

| Paired Comparisons | No. Consumers Responding | | Male Consumers | | Consumer Age (18 to 21 years) | |
|---|---|---|---|---|---|---|
| | Raw | Cooked | Raw | Cooked | Raw | Cooked |
| 0% - 0% | 136 | 114 | 79 | 63 | 110 | 79 |
| 0% - 10% | 111 | 123 | 50 | 70 | 91 | 99 |
| 0% - 20% | 139 | 105 | 87 | 54 | 91 | 64 |
| 0% - 30% | 149 | 104 | 83 | 54 | 79 | 47 |
| 0% - 40% | 105 | 124 | 54 | 71 | 65 | 66 |
| 0% - 50% | 107 | 119 | 56 | 65 | 75 | 76 |
| 0% - 75% | 127 | - | 67 | - | 90 | - |
| 0% - 100% | 100 | - | 50 | - | 63 | - |

Table 2. Recorded consumer responses denoting perceived difference between pairs of noninfested and infested calico scallops when these differences did not exist.

| Recorded Difference | Actual Consumer Responses |
|---|---|
| Appearance | cleaner, dirty, shiney, slimy, artificial, just look different, etc. |
| Color | milky, cloudy, pale, dark, etc. |
| Freshness | spoilage, age, freshness |
| Odor | smell, aroma, odor |
| Shape | uniform, plump, cut, plugs |
| Size | thickness, length, size |
| Texture | smooth, grainy, mushy, firm, soft, fiberous, etc. |

Table 3. Average percent recorded consumer responses for differences which did not exist. Each average is calculated across all eight paired comparisons of infestation per raw and cooked scallops.

| Recorded Response | Average (%) Consumers Responding | |
|---|---|---|
| | Raw Scallops | Cooked Scallops |
| Color | 12.5 | 16.1 |
| Size | 11.2 | 15.1 |
| Appearance | 10.7 | 13.6 |
| Texture | 5.9 | 5.7 |
| Freshness | 5.1 | 3.8 |
| Shape | 4.7 | 5.5 |
| Odor | 1.2 | 1.9 |
| No Difference | 56.1 | 49.6 |

both raw and cooked scallops and regardless of percent infestation compared, differences in color, size and appearance were the most common responses when this difference did not exist (table 3). These 'false' differences were recorded at a higher average response for cooked scallops. Broiling caused a decrease in scallop size from a 130 scallop raw count to a 175 scallop cooked count[2], with a corresponding loss in moisture content from 76.7 percent in raw to 73.7 percent moisture in cooked scallops. The test could not determine if these consequences of cooking influenced consumer perception.

Interestingly, some consumers felt there was a difference in scallop freshness despite the fact that all the scallops had been previously frozen. This result is a reflection on consumer perception and concept of 'freshness' in seafoods. The low average responses for odor as a difference indicates the scallops were above average quality, with no obvious signs of spoilage.

Overall 56.1 percent of the respondents for all the pairs of raw scallops and 49.6 percent for all pairs of cooked scallops found no differences (table 3).

Specific responses recorded as a positive perception of nematodes in scallops are graphed in figure 1. For paired comparisons for raw scallops with one level of infestation less than 50 percent the positive responses ranged from 0 to 1.3 percent perception of nematodes. At 50, 75 and 100 percent levels of infestation 4.7, 8.5 and 20 percent of the respondents gave a positive perception of nematodes, respectively. Thus, under these test conditions consumer perception of nematodes in raw scallops becomes more acute at infestation levels exceeding 50 percent. Sex of the respondent did not influence the results (figure 2).

Cooking seemed to enhance consumer perception of nematodes probably as a result of the muscle shrinkage previously explained. At the 20 and 50 percent level of infestation the percent of positive perception was 2.9 and 10.9 respectively (figure 1). These results would be much lower if the 0 percent infestation level was not available for comparison and/or the cooked scallops were presented as in common practice with sauces and other garnishes.

In conclusion, under the stringent conditions of this study which allowed continuous comparison with 0 percent infestation and encouraged consumer suspicion, the resulting perception of nematodes in calico scallop is apparently low. Perception becomes more acute at infestation levels exceeding 50 percent. Although consumer perception seemed to be enhanced by broiling, the cooked scallops were presented in an atypical fashion without customary sauces,

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[2]Count is a measure of scallop size relative to the total number of individual scallop meats required to constitute one pound.

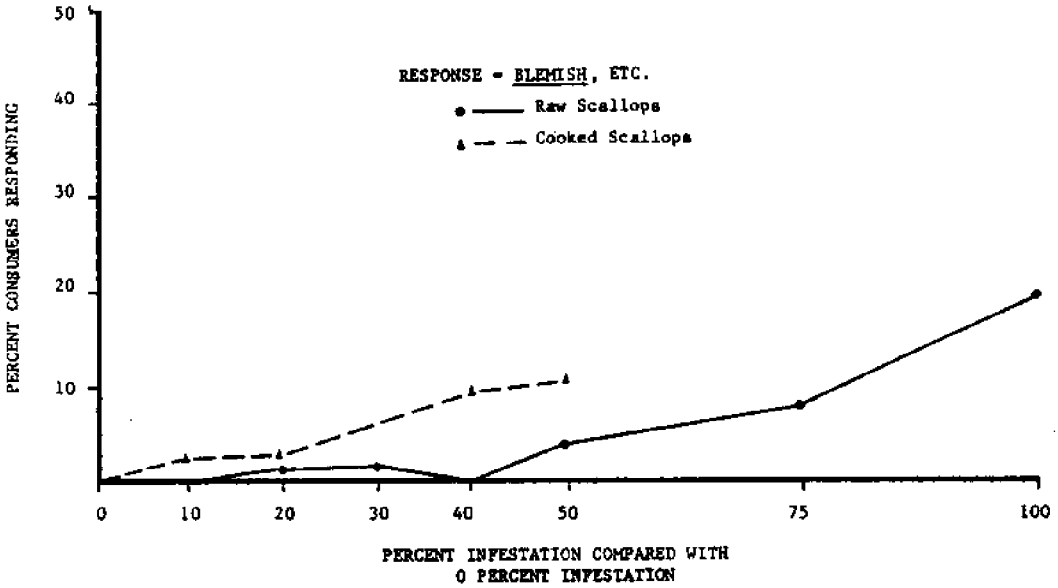


Figure 1. Total percent consumers responding with a positive perception of nematodes denoted in paired comparisons of various percent infestation with 0 percent infestation in raw and cooked scallops.

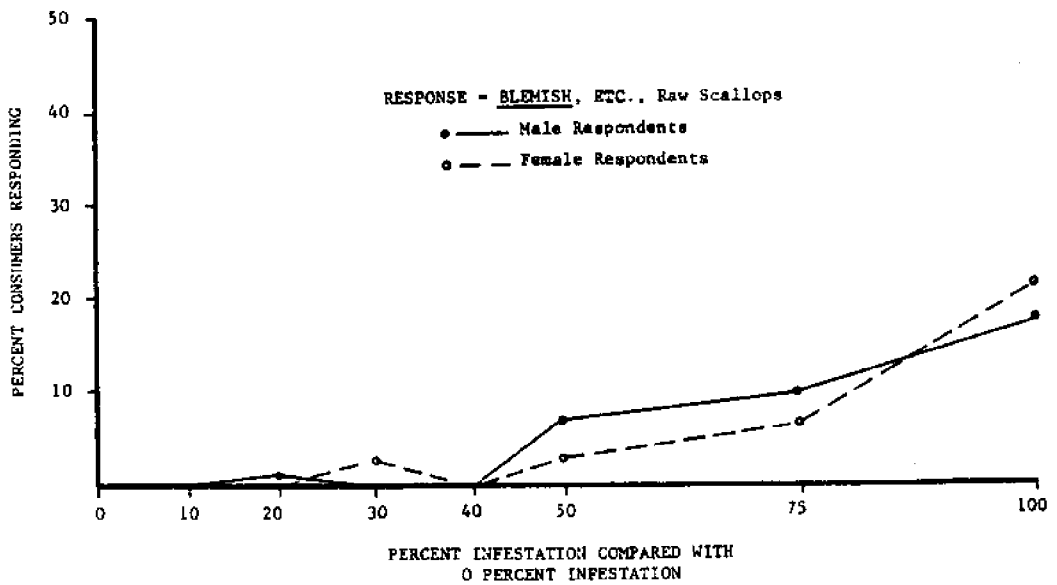


Figure 2. Total percent male and female consumers responding with a positive perception of nematodes denoted in paired comparisons of various percent infestation with 0 percent infestation in raw and cooked scallops.

breadings, garnishes, etc. Consumer responses denoting differences that did not exist can be viewed as a partial measure of suspicion thus,further tempering these results.

## REFERENCES


1. Blake, N.J., B.J. Barber, G.E. Roderick, and C.D. Burns. 1983. Adductor muscle parasites, Sulcascaris sulcata, in calico scallop from the southeast coast of the United States. Proc. Eighth Annual Tropical and Subtropical Fish. Tech. Conf., Tampa, FL.

2. Cobb, N.A. 1930. A nemic parasite of Pecten. J. Parasit. 17: 104-105.

3. Litchtenfels, J.R., T.K. Sawyer, and G.C. Miller. 1980. New hosts for larval Sulcascaris sp. (Nematoda, Anisakidae) and prevalence in the calico scallop (Argopecten gibbus). Trans. Amer. Micros. Soc. 99(4) 448-451.

4. Sawyer, T.K., A. Rosenfield, F.O. Perkins, O.E. Zwerner, R.J. Dias, J.R. Litchtenfels,P.A. Madden, G.J. Jackson, J.W. Bier, W.L. Payne, and G.C. Miller. 1979. Identification of parasitic nematode larvae in the calico scallop, Argopecten gibbus (Linn.) and the surf clam, Spisula solidissma. International Council for the Exploration of the Sea Special Meeting on Diseases of Commercially Important Marine Fish and Shellfish, Report No. 32, 5pp.

# TEXTURE ANALYSIS OF TAIL MEAT FOR MACROBRACHIUM ROSENBERGII

R. Tillman and G. Finne
Department of Animal Science
Texas A&M University
College Station, Texas 77843

## INTRODUCTION

Compared to traditional fisheries, aquaculture is a relatively new form of food production. However, the importance of aquaculture is not at its present level of production but rather in its future potential. With traditional stocks harvested close to their maximum sustainable yield (Moisecv, 1973), much of the future demands for seafoods must be met through aquaculture.

Of many species tested, Malaysian prawn (Macrobrachium rosenbergii) has emerged as an ideal animal for intensive freshwater culture. Freshwater prawns could offer the consumer an attractive alternative to marine shrimp. However, one factor that may limit the development of Macrobrachium rosenbergii is a frequent post-morten texture problem that is associated with these animals. These texture problems, expressed by consumers as "soft", "watery", and "mushy", is presently an obstacle to the successful marketing of freshwater prawns. As pointed out in the National Aquaculture Plan of 1980: "Additional work is required on processing and packaging whole prawns; the 'mushiness' problem also needs attention. Mushiness, coupled with variations of quality and packaging of imported fresh prawns, gives many U.S. wholesalers a negative opinion of prawns."

The objectives of this research were to identify the cause of mushiness that frequently occurs in fresh and frozen Macrobrachium rosenbergii tail muscle, and to develop possible processing methods to reduce or eliminate it.

## MATERIALS AND METHODS

Materials. All freshwater prawns used during this study were grown at Weyerhaueser Company's aquaculture facilities in Homestead, Florida. The shrimp, mixtures of males and females of commercial size (approximately 11 cm), were harvested from the production ponds by seining and placed live in holding tanks until further processing.

### On Site Preparation

Blanching. Prawns, in groups of 25, were removed from holding tanks and placed in a wire basket which was submerged in a water bath kept at 82°C. Blanching times were 30, 60, and 180 seconds, and the internal temperatures of the prawn tail muscles were monitored by thermocouples connected to a Honeywell Multi-Channel Temperature Recorder. After

blanching, the prawns were placed in slush ice to cool, after which half the prawns from each set were frozen, while the other half was kept on ice.

Rupturing the Hepatopancreas. Since preliminary observations had indicated that materials from the hepatopancreas (gut) accelerated tissue softening, the hepatopancreas of a large number of prawns were ruptured before processing. The rupturing was performed immediately after the prawns were removed from the holding tank by cutting the hepatopancreas with a small knife at the tail end. After rupturing of the gut, the prawns were placed on ice to cool, after which half of the prawns were frozen, and the other half put on ice.

Controls. Two sets of controls were used. One set consisted of prawns removed form the holding ponds, cooled and left heads-on, both as frozen and on ice. The second set was treated the same way, but the heads were removed immediately after cooling. The deheaded prawn tails were given a quick rinse before being frozen or put on ice.

Samples from each of the treatments were frozen by placing them on dry ice and in a well-insulated ice chest. The iced samples were repacked in ice and also placed in an ice chest. All samples were airshipped to the Seafood Laboratory at Texas A&M University, College Station, Texas for analysis.

Chemical Analyses

Proteolytic activity. (a) Casein-agar gel diffusion. A casein-agar gel diffusion method as described by Richardson (1970) was initially used to determine proteolytic activity in different regions of both treated and untreated prawns. Gels were prepared according to the procedure of Bjerrum et al. (1975) using a BIO-RAD Protease Detection Kit (Bio-Rad Laboratories). Extracts were prepared from the hepatopancrease and both anterior and posterior tail regions of the prawns by homogenizing these portions for 1 minute, with 3 parts of an aqueous 0.001% Thermsol solution. Thermsol was added in order to inhibit bacterial activity. The extracts were centrifuged at 10,000 rpm for 10 minutes at 4°C and enough supernatant was added to fill 12 mm wells in the casein-agar gel plates. The plates were incubated at 35°C for 24 hours after which the size of the clearing zones was measured.

(b) Spectrophotometric. In order to better quantify proteolytic activity, a colorometric method similar to that described by Lin (1969) was used. This method determines proteolytic activity by the action of enzymes on dimethylcasein.

A 10 ml solution of dimethylcasein was incubated with 0.03 ml of the tissue extracts described above. The reaction was terminated by placing the reaction tubes in boiling water for 60 seconds at regular intervals over a 6 hour time span. The extracts were reacted with trinitrobenzenesulfonic (TBS) acid and the intensity of the yellow chromophore was determined at 340 mm using a Perkin-Elmer Double Beam Spectrophotometer, Model 124.

Sensory Evaluations. Representative numbers of prawns from each treatment were prepared by splitting each prawn longitudinally in half, and broiled on Faberware Open-Hearth broilers for 5 minutes. The prawns were served, while warm, to a trained twelve-member sensory panel and each member independently evaluated each sample for texture (9 = extremely desirable, 1 = extremely undesirable). Panelists were also asked to describe their response using one of the four terms: mushy, soft, firm or tough.

Texture Analysis. Texture analyses were conducted using an Instron Universal Testing Instrument with a 10-blade test cell. Prawn tails were boiled in water for 4 minutes, then cooled on ice until analyzed. Anterior segments of four tails were weighed and placed in the bottom of the shear cell. Five repetitions were performed for each treatment. The ratio of shear force per weight unit was used for statistical analysis.

Statistical Analysis. The effect of different treatments on texture was examined statistically by analysis of variance (ANOVA) and Chi-Square. Duncan's Multiple Range test was used to separate means of the quality attributes.

## RESULTS AND DISCUSSION

Blanching. Blanching had little effect on inactivating proteolysis of prawns held on ice or in frozen storage. All extracts analyzed by casein-agar gel diffusion produced zones of clearing indicating the presence of proteolytic enzymes. Sensory evaluation indicated that the blanched tails had the same texture as the heads-on tails. Interestingly, the 60 second blanch produced larger zones of clearing than those of other blanch periods or than extracts of frozen prawns. This would indicate that the 60 second blanch may have actually enhanced the proteolytic action, and might be detrimental to the texture of the product.

Sensory evaluation of all treatments held on ice indicated little differences in samples after 5 days of treatment with the exception of the heads-off tails. The heads-off tails remained acceptable for 7 days held on ice, while all of the other treatments were judged to have unacceptable texture.

### Results of Analysis of Frozen Samples

Samples were thawed at 4°C for 24 and 48 hours. A third group of samples was thawed by placing under running tap water (0 hour thaw).

### Chemical Analysis

Proteolytic activity. (a) Casein-agar gel diffusion. This test showed proteolytic activity in all extracts prepared from the gut regions; no activity was observed form extracts prepared from extracts prepared from muscle of the posterior region of tails. Extracts prepared from the anterior regions of frozen tails, ruptured and control, produced no zones of clearing in the diffusion gels, indicating no proteolysis. However, extracts from thawed tails of the same sample regions

produced zones of clearing in the gels. These observations indicated that the proteolytic action was possibly being caused by enzymes present in the hepatopancreas that might migrate into the flesh of the tail. Upon thawing, this activity seemed to increase, as noted by larger zones of clearing from extracts of frozen shrimp. This may be caused by cellular disruption during freezing, which may speed the release of the enzymes into solution.

(b) Spectrophotometric. This colorometric method was developed to further quantify the proteolysis in frozen and thawed prawns. Figure 1 presents the results of analyses of extracts prepared from the three regions of frozen and thawed (48 hours) prawns. The gut region of both thawed (48 hours) and unthawed (0 hours) prawns showed proteolytic activity by a measured difference in absorbance. The flattening out of the curve towards the end of the incubation period is thought to be caused as the substrate is expended after long incubation periods. There was no observed differences in absorbance from any of the tail portion of unthawed prawns. The anterior and posterior tail portions of thawed prawns indicated enzymatic activity.

Because the activity in tail portions was unclear in this initial set of colorometric experiments, a second set of analyses was conducted, concentrating on the flesh of thawed prawns (48 hours). Figure 2 shows that there was activity in the anterior tail portion, while there was no activity in the posterior portions.

This could further indicate a migration of the enzymes from the hepatopancreas into the flesh of the tail of frozen prawns. The absorbance noted from the proteolytic activity in the posterior portion is thought to be caused by the reaction of the TBS with the background practices present in the extract.

Sensory Evaluation. Sensory panel members were able to distinguish between samples that were thawed and the sample that had no thaw period (PR F = 0.0239). Examination of the sensory evaluation scores indicated that the thawed samples had a more desirable texture (Table I). However, the Chi-Square analysis of the frequency of the panelists' word description of the texture proved that the 0-hour thawed samples were rated as "tough", while the thawed samples were rated as less firm. Prawns from the three different thaw periods were served at the same time; this may have caused an unconscious comparison of the different samples by the panelists. The preceived toughness may not have been as intense if the samples had been served separately and the toughness would most likely been determined to be a firm tail.

Texture Analysis. The results of the sensory evaluation seem more meaningful when compared with the results of the Instron texture analysis. Texture ration values (shear forces/weight), presented in Table III, also shows a difference between thawed and unthawed samples (PR F = 0.0001). This analysis, like the sensory evaluation, showed that samples taken directly from the freezer and thawed under tap water to be more firm than those thawed at 4°C.

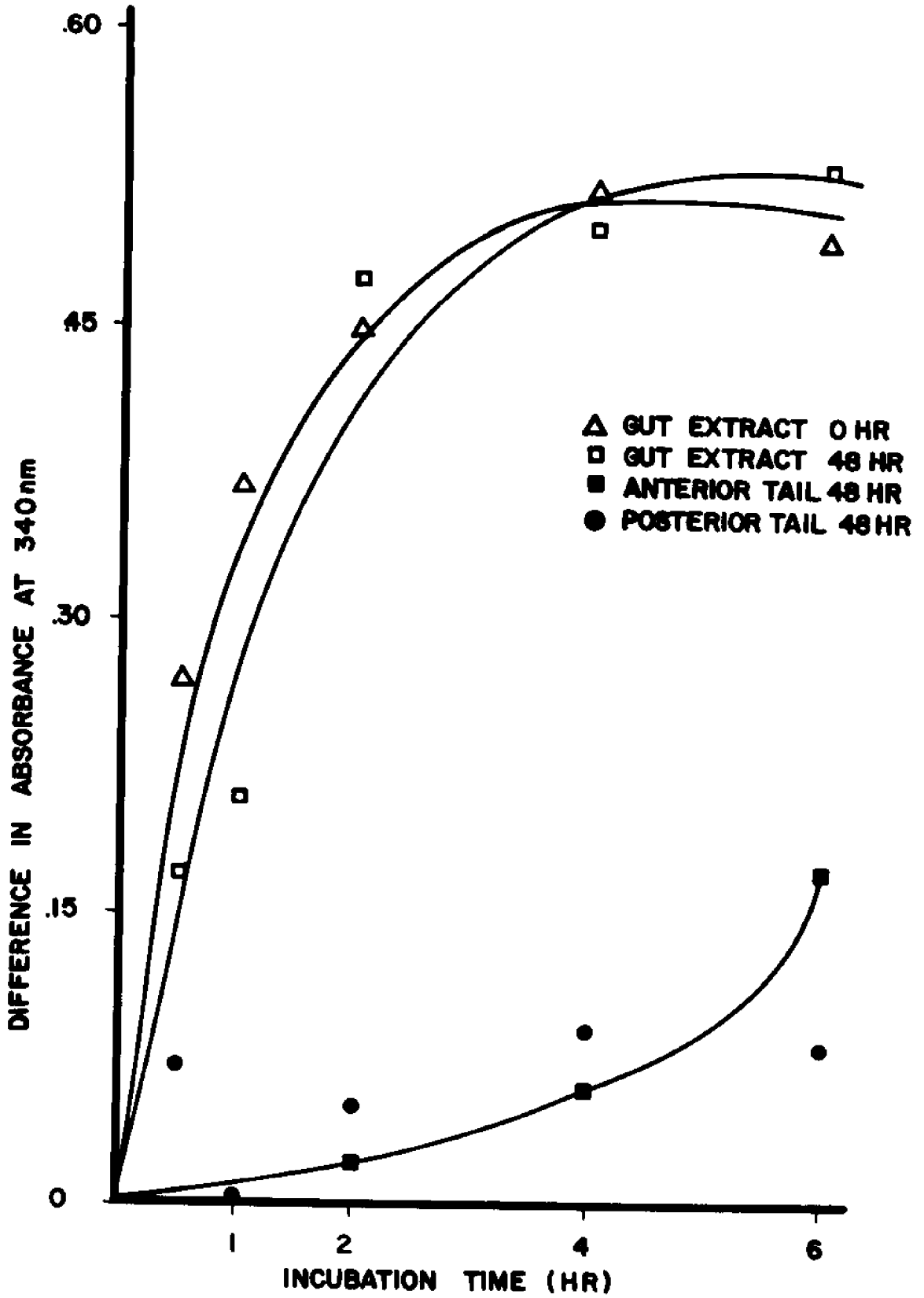


Figure 1. Proteolytic activity of extracts from M. rosenbergii thawed for 48 and 0 hours; 10 ml of dimethylcasein was reacted with 0.03 ml of extract and trinitrobenzene-sulfonic acid.

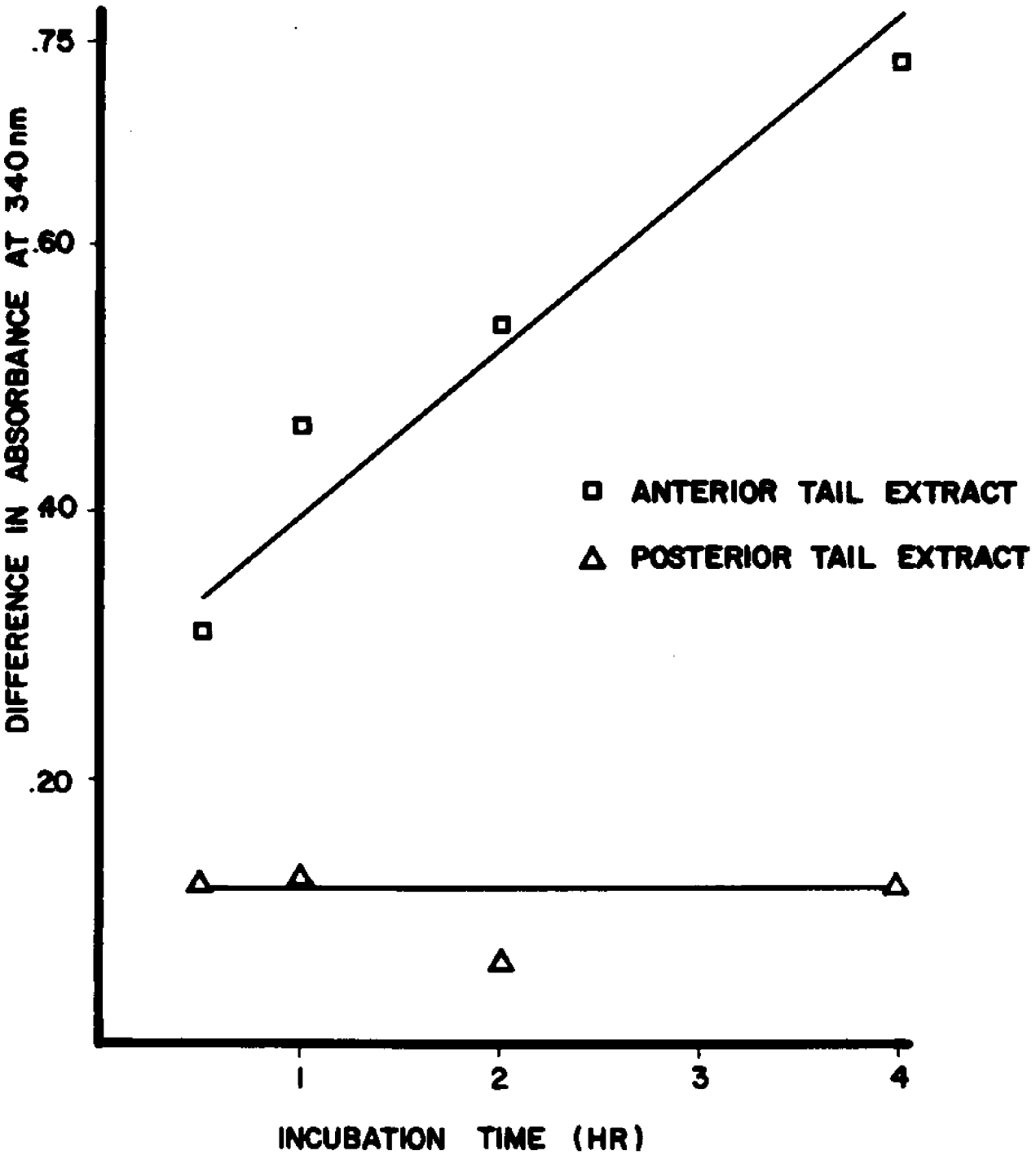


Figure 2. Proteolytic activity of extracts of M. rosenbergii thawed for 48 hours; 10 ml of dimethylcasein was reacted with 0.03 ml of extract and trinitrobenzene-sulfonic acid.

TABLE I. Sensory evaluation scores for frozen M. rosenbergii prepared after different periods of thaw.

| Thaw Period (hr) | Mean Score |
|---|---|
| 24 | 7.3a |
| 48 | 7.0a |
| 0 | 4.9b |

Means followed by the same letter are not significantly different (PR F = 0.0239).

TABLE II. Chi-Square analysis of sensory evaluation scores for M. rosenbergii prepared after different periods of thaw.

| THAW Period | TEXTURE Tough | Firm | Soft | Mushy | |
|---|---|---|---|---|---|
| 24 hr | 0 | 9 | 1 | 1 | Frequency |
| | 3.0 | 6.7 | 1.3 | 1.0 | Expected |
| 48 hr | 1 | 7 | 3 | 2 | Frequency |
| | 3.0 | 6.7 | 1.3 | 1.0 | Expected |
| 0 hr | 8 | 4 | 0 | 0 | Frequency |
| | 3.0 | 6.7 | 1.3 | 1.0 | Expected |
| | | prob. = 0.027 | | | |

TABLE III. Allo-Kramer Shear Force/Weight Ratio values of 4 anterior segments of frozen M. rosenbergii prepared after different periods of thaw.

| Thaw Period (hr) | Ratio Mean |
|---|---|
| 0 | 9.9a |
| 48 | 7.1b |
| 24 | 6.8b |

Means followed by the same letter are not significantly different (PR F = 0.0001).

## CONCLUSION

Research on the texture of tails from Macrobrachium rosenbergii has shown that:

1. Tails of Macrobrachium rosenbergii stored heads-on on ice or refrigerated will become soft and mushy. These texture problems will render the prawn tails unpalatable. Tails from prawns deheaded immediately after harvesting are much more stable and have limited texture problems.
2. The development of mushiness was progressive from the anterior to the posterior portion of the tail. Upon prolonged storage, the mushiness gradually increased.
3. When stored on ice, properly harvested prawns tended to develop softness by the head segments after two days on ice.
4. Frozen prawns stored with heads-on did not develop significant softness until thawed. Upon thawing, these prawns developed mushiness faster than fresh prawns stored on ice immediately after harvesting.
5. The causative agent for mushiness development in tails of freshwater prawns seems to be the action of proteolytic enzymes on the muscle fibers of the prawn tail. Since this proteolytic action is limited to prawns stored heads-on, it is more likely that the causative agents are released from the head portion of the prawn. This is supported by the fact that the softening gradually spreads from the anterior part of the prawn tail towards the posterior.
6. In order to try to inhibit proteolysis, prawns were heat-blanched for different time intervals. Blanching shrimp at 82 C for three minutes had little or no effect on development of soft tails. In order to effectively inhibit proteolysis by heat, a treatment is required which would result in the surface of the prawn becoming cooked before an inhibition temperature in the deep muscle is reached. Therefore, blanching is not recommended.

## ACKNOWLEDGEMENTS

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## REFERENCES

Bjerrum, O.J. 1975. Quantitative Immunoelectrophoresis. Scandinavian J. Immunology, Supplement 2, N.H. Axelsen, ed.:82-83.

Lin, Y., Means, G.E., and Fenney, R.E. 1969. The Action of Proteolytic Enzymes on NN-Bimethyl Proteins. JBC, 244:789-793.

Moislev, P.A. 1973. Development of Fisheries for Traditionally Exploited Species. J. Fish. Res. Bal. Canada. 30:2109-2120.

Richardson, G.H. 1970. Differentiation of Enzyme Coagulants on Casein-Agar gels.